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THE RESOURCES AGENCY
DEPARTMENT OF FISH AND GAME

HAZARD ASSESSMENT OF THE INSECTICIDE CHLORPYRIFOS TO AQUATIC ORGANISMS IN THE SACRAMENTO-SAN JOAQUIN RIVER SYSTEM

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PREFACE

The California Department of Fish and Game (CDFG) is responsible for protection and management of fish and wildlife. The CDFG protects fish and wildlife from pesticide hazards through consultation with the California Environmental Protection Agency's Department of Pesticide Regulation (DPR) Pesticide Registration and Evaluation Committee and Pesticide Advisory Committee. The State Water Resources Control Board and the Regional Water Quality Control Boards also protect fish and wildlife by promulgating and enforcing water quality standards for pesticides and other toxic materials. In recognition of the need for applicable environmental standards for fish and wildlife, DPR contracted with CDFG to assess the effects of pesticides on fish and wildlife and to facilitate the development of water quality criteria to protect aquatic organisms.

This document is the fourth in a series of pesticide hazard assessments. Hazard assessments have also been prepared for the herbicides molinate and thiobencarb and the insecticides methyl parathion and carbofuran.

Hazard Assessment of the Insecticide Chlorpyrifos to Aquatic Organisms in the Sacramento-San Joaquin River System

by

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SUMMARY

Interim freshwater and final saltwater Water Quality Criteria (WQC) for protection of aquatic organisms from the insecticide chlorpyrifos were developed and a hazard assessment was performed for California's Sacramento-San Joaquin River system.

One hundred twenty tests on the acute and chronic toxicity of chlorpyrifos to aquatic organisms were reviewed and evaluated. The most acutely sensitive freshwater species tested was the cladoceran *Ceriodaphnia dubia* with a mean 96-h LC_{50} value of 0.10 µg/L. Maximum Acceptable Toxicant Concentration (MATC) values were available for only one freshwater species, the fathead minnow *Pimephales promelas*; the mean MATC value was 3.44 µg/L. The most acutely and chronically sensitive saltwater species tested was the mysid *Mysidopsis bahia* with a mean LC_{50} value of 0.04 and MATC value of 0.003 µg/L.

The calculated freshwater Final Acute Value (FAV) for chlorpyrifos was 0.07 µg/L. The calculated saltwater FAV was 0.03 µg/L. The Final Acute-Chronic Ratio (FACR) for both freshwater and saltwater was 4. The freshwater Final Chronic Value (FCV) was 0.02 µg/L (FCV=FAV/FACR). The saltwater Final Chronic Value (FCV) was 0.01 µg/L. Interim freshwater and final saltwater WQC of 0.02 and 0.01 µg/L, respectively, are proposed for the Sacramento-San Joaquin Estuary. The U.S. Environmental Protection Agency (EPA) freshwater and saltwater chronic WQC are 0.041 and 0.0056 µg/L, respectively. The DFG-proposed criteria are similar to the EPA criteria in that they differ by a factor of two or less.

Chlorpyrifos monitoring data are not available for the Sacramento River system. Detected concentrations in the San Joaquin River system ranged from 0.01 to 1.6 µg/L from March 1991 through February 1993. Concentrations greater than the WQC of 0.02 µg/L have been detected in the San Joaquin River system for periods of time long enough to result in chronic exposure for aquatic organisms.

Chronic toxicity data for freshwater invertebrates, particularly *Ceriodaphnia dubia*, are necessary to better define the FACR. Additional chronic toxicity data for species for which acute data are available would also be useful.

Monitoring of the Sacramento-San Joaquin River system should be continued and improved to include daily concentrations during several months to better assess the hazard posed by chlorpyrifos to aquatic species.

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ACKNOWLEDGMENTS

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INTRODUCTION

The organophosphate insecticide chlorpyrifos is applied to alfalfa, citrus, cotton, apples, nuts, vegetables, and other crops. In 1990, the only year for which complete data are available, 1,120,364 kg of chlorpyrifos were used in California (California Department of Pesticide Regulation [DPR] 1990).

Chlorpyrifos was detected periodically in the San Joaquin River system by the Central Valley Regional Water Quality Control Board (CVRWQCB) and the California Department of Pesticide Regulation (DPR) from March 1991 through February 1993 (Table 1). Detected concentrations ranged from 0.01 µg/L to 1.6 µg/L. The U.S. Geological Survey (USGS) monitors the Sacramento River system for chlorpyrifos, but data are not yet available. No chlorpyrifos monitoring data were available for saltwater.

The toxic effects of chlorpyrifos on aquatic animals were assessed by evaluating toxicity tests published in the scientific literature and corporate laboratory reports from confidential files submitted to DPR in support of pesticide registration. Chlorpyrifos toxicity tests were evaluated for conformance with specific criteria adapted from U.S. Environmental Protection Agency (EPA) (1985) guidelines and the American Society of Testing and Materials (ASTM) (1978, 1980, 1983, 1984, 1985, 1987, 1988a, 1988b, 1988c, 1989, 1990, 1992). Although toxicity tests were not required to comply with all criteria, tests were rejected if they did not observe important procedures such as maintaining sufficient organism survival in control treatments. The Water Quality Criterion (WQC) was calculated using data from accepted tests and methods adapted from EPA (1985) guidelines (Appendix D).

Table 1. Concentrations of chlorpyrifos (µg/L) detected in the San Joaquin River system, March 1991 through February 1993.

Date	Location[a]	Concentration
4/13/92	Bishop Cut at Eight Mile Road	0.01[b]
3/19/91	Del Puerto Creek	0.12[b]
4/23/91	Del Puerto Creek	0.04[b]
5/15/91	Del Puerto Creek	0.02[b]
5/28/91	Del Puerto Creek	0.01[b]
12/23/91	Del Puerto Creek	0.01[b]
1/05/92	Del Puerto Creek	0.01[b]
2/03/92	Del Puerto Creek	0.01[b]
2/10/92	Del Puerto Creek	0.03[b]
4/27/92	Del Puerto Creek	0.03[b]
5/04/92	Del Puerto Creek	0.02[b]
5/11/92	Del Puerto Creek	0.04[b]
5/18/92	Del Puerto Creek	0.01[b]
5/25/92	Del Puerto Creek	0.01[b]
6/01/92	Del Puerto Creek	0.02[b]
6/15/92	Del Puerto Creek	0.01[b]
6/22/92	Del Puerto Creek	0.04[b]
2/08/93	Highline Spillway	0.07[c]
3/19/91	Ingram/Hospital Creeks	0.57[b]
4/23/91	Ingram/Hospital Creeks	0.03[b]
5/15/91	Ingram/Hospital Creeks	0.01[b]
5/28/91	Ingram/Hospital Creeks	0.02[b]
9/09/91	Ingram/Hospital Creeks	0.33[b]
1/28/92	Ingram/Hospital Creeks	0.01[b]
2/10/92	Ingram/Hospital Creeks	0.01[b]
3/09/92	Ingram/Hospital Creeks	0.01[b]
3/16/92	Ingram/Hospital Creeks	0.06[b]
4/27/92	Ingram/Hospital Creeks	0.01[b]
5/04/92	Ingram/Hospital Creeks	0.02[b]
5/11/92	Ingram/Hospital Creeks	0.01[b]
5/18/92	Ingram/Hospital Creeks	0.01[b]
5/25/92	Ingram/Hospital Creeks	0.01[b]
6/15/92	Ingram/Hospital Creeks	0.01[b]
6/22/92	Ingram/Hospital Creeks	0.01[b]
2/08/93	Livingston Spillway	0.10[c]
4/23/91	Los Banos Creek	0.01[b]
4/23/91	Merced River	0.01[b]
1/20/92	Merced River	0.01[b]
1/28/92	Merced River	0.02[b]
2/17/92	Merced River	0.05[b]
2/18/92	Merced River	0.06[c]
3/09/92	Merced River	0.01[b]
4/13/92	Merced River	0.13[b]
4/27/92	Merced River	0.01[b]
5/04/92	Merced River	0.01[b]
2/07/93	Merced River	0.07[c]
2/09/93	Merced River	0.06[c]

Table 1. Continued -2-

Date	Location[a]	Concentration
4/23/91	Mud Slough	0.01[b]
4/23/91	Newman Wasteway	0.01[b]
1/15/93	Newman Wasteway	0.12[c]
2/09/93	Newman Wasteway	0.14[c]
3/19/91	Orestimba Creek	0.12[b]
4/18/91	Orestimba Creek	0.15[b]
4/23/91	Orestimba Creek	0.01[b]
5/15/91	Orestimba Creek	0.12[b]
7/30/91	Orestimba Creek	0.72[b]
2/10/92	Orestimba Creek	0.02[b]
4/20/92	Orestimba Creek	0.02[b]
4/27/92	Orestimba Creek	0.09[b]
5/04/92	Orestimba Creek	0.08[b]
5/11/92	Orestimba Creek	0.02[b]
5/18/92	Orestimba Creek	0.01[b]
5/25/92	Orestimba Creek	0.01[b]
6/15/92	Orestimba Creek	0.01[b]
6/22/92	Orestimba Creek	0.02[b]
3/16/92	Salt Slough at HWY 165	0.01[b]
4/13/92	Salt Slough at HWY 165	0.12[b]
5/15/91	San Joaquin River at Airport Road	0.01[b]
1/20/92	San Joaquin River at Airport Road	0.01[b]
1/28/92	San Joaquin River at Airport Road	0.03[b]
2/17/92	San Joaquin River at Airport Road	0.02[b]
3/09/92	San Joaquin River at Airport Road	0.03[b]
4/27/92	San Joaquin River at Airport Road	0.01[b]
5/04/92	San Joaquin River at Airport Road	0.01[b]
5/11/92	San Joaquin River at Airport Road	0.01[b]
4/23/91	San Joaquin River at Fremont Ford Park	0.01[b]
1/28/92	San Joaquin River at Fremont Ford Park	0.01[b]
2/17/92	San Joaquin River at Bowman Road	0.01[b]
6/12/91	San Joaquin River at Hills Ferry Road	0.01[b]
1/28/92	San Joaquin River at Hills Ferry Road	0.02[b]
3/16/92	San Joaquin River at Hills Ferry Road	0.01[b]
4/27/92	San Joaquin River at Hills Ferry Road	0.01[b]
5/11/92	San Joaquin River at Hills Ferry Road	0.02[b]
3/18/91	San Joaquin River at Laird Park	0.05[c]
4/25/91	San Joaquin River at Laird Park	0.05[c]
4/26/91	San Joaquin River at Laird Park	0.07[b]
5/15/91	San Joaquin River at Laird Park	0.01[b]
5/28/91	San Joaquin River at Laird Park	0.02[b]
1/20/92	San Joaquin River at Laird Park	0.01[b]
1/28/92	San Joaquin River at Laird Park	0.02[b]
2/03/92	San Joaquin River at Laird Park	0.01[b]
3/09/92	San Joaquin River at Laird Park	0.04[b]

Table 1. Continued -3-

Date	Location[a]	Concentration
3/16/92	San Joaquin River at Laird Park	0.01[b]
3/30/92	San Joaquin River at Laird Park	0.01[b]
4/13/92	San Joaquin River at Laird Park	0.02[b]
4/20/92	San Joaquin River at Laird Park	0.03[b]
4/27/92	San Joaquin River at Laird Park	0.02[b]
5/04/92	San Joaquin River at Laird Park	0.02[b]
5/11/92	San Joaquin River at Laird Park	0.02[b]
5/18/92	San Joaquin River at Laird Park	0.01[b]
6/01/92	San Joaquin River at Laird Park	0.01[b]
6/15/92	San Joaquin River at Laird Park	0.01[b]
6/22/92	San Joaquin River at Laird Park	0.01[b]
8/19/92	San Joaquin River at Laird Park	0.35[c]
2/10/93	San Joaquin River at Laird Park	0.06[c]
2/11/93	San Joaquin River at Laird Park	0.07[c]
4/23/91	San Joaquin River at Maze Blvd	0.02[b]
1/28/92	San Joaquin River at Maze Blvd	0.01[b]
4/25/91	San Joaquin River at Patterson	0.08[c]
2/10/93	San Joaquin River at Patterson	0.08[c]
4/26/91	San Joaquin River at West Main	0.09[b]
1/28/92	San Joaquin River at West Main	0.01[b]
3/19/91	Spanish Grant Combined Drain	0.47[b]
4/18/91	Spanish Grant Combined Drain	0.11[b]
5/15/91	Spanish Grant Combined Drain	0.22[b]
5/28/91	Spanish Grant Combined Drain	0.21[b]
6/12/91	Spanish Grant Combined Drain	0.03[b]
2/10/92	Spanish Grant Combined Drain	0.08[b]
4/27/92	Spanish Grant Combined Drain	0.19[b]
5/04/92	Spanish Grant Combined Drain	0.07[b]
5/11/92	Spanish Grant Combined Drain	0.04[b]
5/18/92	Spanish Grant Combined Drain	0.05[b]
5/25/92	Spanish Grant Combined Drain	0.03[b]
6/01/92	Spanish Grant Combined Drain	0.17[b]
6/15/92	Spanish Grant Combined Drain	0.01[b]
6/22/92	Spanish Grant Combined Drain	0.01[b]
4/23/91	Stanislaus River	0.01[b]
6/12/91	Stanislaus River	0.01[b]
1/28/92	Stanislaus River	0.01[b]
5/11/92	Stanislaus River	0.01[b]
6/22/92	Stanislaus River	0.01[b]
1/20/92	Tuolumne River	0.02[b]
1/28/92	Tuolumne River	0.01[b]
2/17/92	Tuolumne Road	0.03[b]
4/27/92	Tuolumne River	0.01[b]
5/04/92	Tuolumne River	0.01[b]
5/11/92	Tuolumne River	0.01[b]
5/25/92	Tuolumne River	0.01[b]
6/15/92	Tuolumne River	0.01[b]

Table 1. Continued -4-

Date	Location[a]	Concentration
3/04/91	Turlock Irrigation Drain #3	0.12[b]
3/19/91	Turlock Irrigation Drain #3	0.23[b]
4/04/91	Turlock Irrigation Drain #3	0.06[b]
1/05/92	Turlock Irrigation Drain #3	0.02[b]
1/20/92	Turlock Irrigation Drain #3	1.60[b]
2/10/92	Turlock Irrigation Drain #3	0.74[b]
2/17/92	Turlock Irrigation Drain #3	0.17[b]
3/02/92	Turlock Irrigation Drain #3	0.04[b]
3/09/92	Turlock Irrigation Drain #3	0.12[b]
3/16/92	Turlock Irrigation Drain #3	0.04[b]
5/04/92	Turlock Irrigation Drain #3	0.01[b]
5/11/92	Turlock Irrigation Drain #3	0.01[b]
5/18/92	Turlock Irrigation Drain #3	0.01[b]
3/19/91	Turlock Irrigation Drain #5	0.05[b]
4/04/91	Turlock Irrigation Drain #5	0.02[b]
4/25/91	Turlock Irrigation Drain #5	0.23[c]
4/26/91	Turlock Irrigation Drain #5	0.19[b]
12/18/91	Turlock Irrigation Drain #5	0.01[b]
1/05/92	Turlock Irrigation Drain #5	0.01[b]
1/13/92	Turlock Irrigation Drain #5	0.01[b]
1/28/92	Turlock Irrigation Drain #5	0.01[b]
2/03/92	Turlock Irrigation Drain #5	0.01[b]
2/10/92	Turlock Irrigation Drain #5	0.04[b]
2/17/92	Turlock Irrigation Drain #5	0.08[b]
3/09/92	Turlock Irrigation Drain #5	0.08[b]
4/27/92	Turlock Irrigation Drain #5	0.02[b]
5/04/92	Turlock Irrigation Drain #5	0.01[b]
5/11/92	Turlock Irrigation Drain #5	0.05[b]
5/25/92	Turlock Irrigation Drain #5	0.01[b]
6/01/92	Turlock Irrigation Drain #5	0.01[b]
6/22/92	Turlock Irrigation Drain #5	0.01[b]
2/09/93	Turlock Irrigation Drain #5	0.07[c]
5/28/91	Turlock Irrigation Drain #6	0.15[b]
7/15/91	Turlock Irrigation Drain #6	0.01[b]
1/20/92	Turlock Irrigation Drain #6	0.17[b]
1/20/92	Turlock Irrigation Drain #6	0.01[b]
2/03/92	Turlock Irrigation Drain #6	0.05[b]
2/10/92	Turlock Irrigation Drain #6	0.12[b]
2/17/92	Turlock Irrigation Drain #6	0.04[b]
4/06/92	Turlock Irrigation Drain #6	0.14[b]
4/27/92	Turlock Irrigation Drain #6	0.01[b]
5/04/92	Turlock Irrigation Drain #6	0.01[b]
5/11/92	Turlock Irrigation Drain #6	0.07[b]
6/01/92	Turlock Irrigation Drain #6	0.25[b]
6/22/92	Turlock Irrigation Drain #6	0.01[b]

[a] These and other locations were sampled in 1991, 1992, and 1993. Only dates on which chlorpyrifos was detected are listed.

[b] Unpublished data from monitoring by Central Valley Regional Water Quality Control Board.

[c] Unpublished data from monitoring by Department of Pesticide Regulation.

ENVIRONMENTAL FATE

Chlorpyrifos is an organophosphate insecticide. Johnson (1991) classified chlorpyrifos as a non-leacher with water solubility of 0.71 ppm, hydrolysis $t_{1/2}$ of 73 days, and K_{oc} of 17000 cm^3/gm. Chlorpyrifos adsorbs to organic material, and applied chlorpyrifos has been found to remain within one to five inches of the soil surface (Bidlack 1979). McCall (1985) found that 79 to 96% of applied chlorpyrifos remained within the top centimeter of soil.

Because of chlorpyrifos's tendency to adsorb to soil, its bioavailability has been found to be fairly low. Chlorpyrifos is rapidly metabolized by aquatic organisms and biomagnification has not been observed. (Racke 1993)

Because of its low solubility and high adsorption chlorpyrifos has a low potential for runoff in moving water. Movement off agricultural fields has been shown to occur, however, with chlorpyrifos adsorbed to runoff sediments. Chlorpyrifos dissipates rapidly from the water column ($t_{1/2}$ value <24 hours); dissipation from sediments is similar to soil ($t_{1/2}$ value 1 to 16 days). (Racke 1993)

In two soils used in growing rice and under mixed aerobic and anaerobic conditions, chlorpyrifos $t_{1/2}$ values were 15 days and 58 days, respectively. In the same two soils, under anaerobic conditions, chlorpyrifos $t_{1/2}$ values were 39 days and 51 days, respectively (Bidlack 1979). Chlorpyrifos applied to sandy loam and organic soils had $t_{1/2}$ values of <1 week and 2.5 weeks, respectively (Verschueren 1983).

From the information on soil half-life, it appears that under certain soil conditions chlorpyrifos may be persistent in soil. Under those conditions, chlorpyrifos could be expected to be a chronic contaminant of agricultural runoff water and perhaps river systems.

ACUTE TOXICITY TO AQUATIC ANIMALS

One hundred and nine tests on the acute toxicity of chlorpyrifos to aquatic animals were evaluated and abstracted (Appendix A). Seventy of these tests were accepted (Table A-1) and 39 were not accepted (Table A-2). Reasons for rejection are included in the abstracts and in Table A-2. EPA (1985) guidelines recommend eight families of freshwater organisms for which data should be available for deriving a freshwater Final Acute Value (FAV), and eight families of saltwater organisms for deriving a saltwater FAV (Tables 2 and 3). Acceptable tests were available for all of the eight freshwater and saltwater families of organisms (Tables 2 and 3).

EPA (1985) guidelines were used to calculate a freshwater FAV and a saltwater FAV using Genus Mean Acute Values (GMAVs). The GMAVs were derived from all available and acceptable Species Mean Acute Values (SMAVs) for species in that genus, and the SMAVs were derived from all available and acceptable acute values for that species. The GMAVs were ranked in ascending order (Tables 4 and 5). Freshwater GMAVs ranged from 0.10 µg/L, the mean 96-h LC_{50} value for the cladoceran *Ceriodaphnia dubia*, to >806 µg/L, the 96-h LC_{50} value for both the goldfish *Carassius auratus* and the snail *Apnexa hypnorum*. Saltwater GMAVs ranged from 0.04 µg/L, the mean 96-h LC_{50} value for the mysid *Mysidopsis bahia*, to 1991 µg/L, the 96-h LC_{50} value for the eastern oyster *Crassostrea virginica*.

The four lowest GMAVs are the most significant determinants of the FAVs. For the freshwater FAV the lowest four GMAVs were for invertebrate species. For the saltwater FAV three of the four lowest GMAVs were for invertebrate species. The calculated freshwater and saltwater FAVs were 0.07 µg/L and 0.03 µg/L, respectively.

Table 2. Eight families of freshwater aquatic animals represented in the Final Acute Value.

Family	Animal Used
1. One Salmonid	Rainbow trout
2. Another family in class Osteichthyes	Fathead minnow
3. Another family in phylum Chordata	Bluegill
4. One family not in phylum Arthropoda or Chordata	Amphipod
5. One insect family or any phylum not already represented	Stonefly
6. One planktonic crustacean	*Ceriodaphnia dubia*
7. One benthic crustacean	Crayfish
8. One insect	Crawling water beetle

Table 3. Eight families of saltwater aquatic animals represented in the Final Acute Value.

Family	Animal Used
1, 2. Two families in phylum Chordata	Silverside, striped mullet
3. One family not in phylum Arthropoda or Chordata	Eastern oyster
4, 5, 6. Three other families not in phylum Chordata	Brown shrimp, blue crab, *Mysidopsis bahia*
7. A mysid or penaeid	Pink shrimp
8. One other family not already represented	California grunion

Table 4. Ranked Genus Mean Acute Values (GMAV) from accepted acute toxicity tests on freshwater species used to calculate the freshwater FAV.

Rank	GMAV[a] (µg/L)	Species
1	0.10 Cladoceran	*Ceriodaphnia dubia*
2	0.11	Amphipod *Gammarus lacustris*
3	0.15 Mysid[b]	*Neomysis mercedis*
4	0.38	Stonefly *Pteronarcella badia*
5	0.57	Stonefly *Claassenia sabulosa*
6	0.80	Crawling water beetle *Petodytes sp.*
7	1.0	Cladoceran *Daphnia magna*
8	3.03	Bluegill[b] *Lepomis macrochirus*
9	6.0	Crayfish *Orconectes immunis*
10	10	Stonefly *Pteronarcys californica*
11	10.4	Rainbow trout[b] *Oncorhynchus mykiss* Cutthroat trout *Oncorhynchus clarki*
12	244	Lake trout *Salvelinus namaycush*
13	249	Fathead minnow[b] *Pimephales promelas*
14	475	Channel catfish[b] *Ictalurus punctatus*
15	>806	Goldfish[b] *Carassius auratus*
16	>806	Snail *Aplexa hypnorum*

Freshwater FAV: 0.07

[a] Represents Species Mean Acute Value (SMAV) if only one species is listed. Individual values are listed in Table A-1.

[b] Occurs in Sacramento-San Joaquin River system.

Table 5. Ranked Genus Mean Acute Values (GMAV) from accepted acute toxicity tests on saltwater species used to calculate the saltwater FAV.

Rank	GMAV[a] (µg/L)	Species
1	0.04	Mysid *Mysidopsis bahia*
2	0.69	Brown shrimp *Penaeus aztecus* Pink shrimp *Penaeus duorarum*
3	1.2	California grunion *Leuresthes tenuis*
4	1.5	Grass shrimp *Palaemonetes pugio*
5	1.5	Atlantic silverside[b] *Menidia menidia* Tidewater silverside *Menidia peninsulae* Inland silverside *Menidia beryllina*
6	2.7	Gulf killifish *Fundulus grandis* Longnose killifish *Fundulus similis*
7	5.2	Blue crab *Callinectes sapidus*
8	5.4	Striped mullet *Mugil cephalus*
9	7.0	Spot *Leiostomus xanthurus*
10	188	Gulf toadfish *Opsanus beta*
11	194	Sheepshead minnow *Cyprinodon variegatus*
12	1991	Eastern oyster *Crassostrea virginica*

Saltwater FAV: 0.03

[a] Represents Species Mean Acute Value (SMAV) if only one species is listed. Individual values are listed in Table A-1.

[b] Occurs in Sacramento-San Joaquin River system.

CHRONIC TOXICITY TO AQUATIC ANIMALS

Eleven tests on the chronic toxicity of chlorpyrifos were evaluated for use in deriving the Final Chronic Value (FCV) (Appendix B). Seven of these tests were accepted (Table B-1); four were not accepted (Table B-2). Of the seven accepted tests, only two tests were for a freshwater species, the fathead minnow *Pimephales promelas*.

The No Observable Effect Concentration (NOEC) values from acceptable tests ranged from 0.002 µg/L to 3.7 µg/L chlorpyrifos for a 28-d exposure and a 7-d exposure to mysid *Mysidopsis bahia* and fathead minnow, respectively (Table B-1). The Lowest Observable Effect Concentration (LOEC) value ranged from 0.004 µg/L to 7.4 µg/L for mysid *M. bahia* and fathead minnow, respectively (Table B-1). The Maximum Acceptable Toxicant Concentration (MATC) [$(NOEC \times LOEC)^{1/2}$] ranged from 0.003 µg/L for mysid *M. bahia* to 5.23 µg/L for fathead minnow (Table B-1).

The EPA (1985) guidelines specify calculating the Acute-Chronic Ratio (ACR) for a species using for the numerator the geometric mean of LC_{50} values and for the denominator the geometric mean of MATC values. Additionally, these acute and chronic values should be from the same study. However, insufficient data were available to use this method of deriving all ACR values. Instead, the ACR value was calculated for each species having acute and chronic toxicity data, even though the data were not from the same study. The ACR value was obtained by dividing the geometric mean of all available LC_{50} values from accepted tests by the available MATC value from accepted chronic tests. When an LC_{50} value and an MATC value from the same study were available, as for the fathead minnow, these values were kept separate from the other values for that species and a separate ACR value was calculated (Table 6).

The EPA (1985) guidelines specify that freshwater or saltwater Final Acute to Chronic Ratio (FACR) values be derived using ACR values of both freshwater and saltwater species, including at least a fish, an invertebrate, and an acutely sensitive species. The FACR value used to derive a freshwater FCV should include an acutely sensitive freshwater species. The FACR value used to derive a saltwater FCV should include an acutely sensitive saltwater species. The other species used may be either freshwater or saltwater. ACR values were available for a sufficient range of saltwater species to derive a saltwater FACR value using the guidelines. However, the only freshwater species for which an ACR value was available was the fathead minnow, which is not an acutely sensitive species. Because the EPA (1985) guidelines allow chronic data from freshwater and saltwater species to be combined, the saltwater FACR value was used as the FACR value for both freshwater and saltwater FCVs. It was considered less desirable to use an FACR value based on only one freshwater organism, the fathead minnow, that is not acutely sensitive.

In addition, the EPA (1985) guidelines specify that if the ACR values increase or decrease as the SMAVs increase, as the ACR values generally do with chlorpyrifos, only species with SMAVs close to the FAV should be used to calculate the FACR. Accordingly, the FACR value for chlorpyrifos was calculated as the geometric mean of ACR values for the mysid *Mysidopsis bahia,* and two fish, the tidewater silverside *Menidia peninsulae* and the inland silverside *M. beryllina* (Table 5). The mysid is an acutely sensitive invertebrate. Tidewater and inland silversides are both fish; the tidewater silverside is acutely sensitive.

Using the methods described above, the resultant FACR value is 4. The freshwater FCV is 0.02 µg/L (FAV 0.07/FACR 4); the saltwater FCV is 0.01 µg/L (FAV 0.03/FACR 4).

Table 6. Acute-Chronic Ratio (ACR) Values for freshwater and saltwater species for which acute and chronic toxicity data were available.

Species	LC_{50} or SMAV (µg/L)	NOEC LOEC (µg/L)	MATC $(NOEC \times LOEC)^{1/2}$ (µg/L)	ACR (LC_{50}/MATC)
Mysid *Mysidopsis bahia*	0.040[a]	NOEC 0.002 LOEC 0.004	0.003	13.3[b]
Tidewater silverside *Menidia peninsulae*	0.71[a]	NOEC 0.38 LOEC 0.78	0.54	1.3[b]
Inland silverside *Menidia beryllina*	4.2	NOEC 0.75 LOEC 1.8	1.16	3.6[b]
Fathead minnow *Pimephales promelas*	140[c]	NOEC 1.6 LOEC 3.2	2.26	61.9
Sheepshead minnow *Cyprinodon variegatus*	194[a]	NOEC 1.7 LOEC 3.0	2.26	85.8
Fathead minnow *Pimephales promelas*	249[c]	NOEC 3.7 LOEC 7.4	5.23	47.6
Gulf toadfish *Opsanus beta*	520[c]	NOEC 1.4 LOEC 3.7	2.28	228

Final Acute-Chronic Ratio: 4

[a] Species Mean Acute Value: geometric mean of values from several tests on this species. Individual values are listed in Table A-1.

[b] ACR value used to calculate Final ACR value.

[c] LC_{50} value and MATC value from same test.

TOXICITY TO AQUATIC PLANTS

Seven tests on the toxicity of chlorpyrifos to aquatic plants and bacteria were evaluated (Appendix C) to derive a Final Plant Value (FPV). The FPV is the lowest concentration of pesticide that demonstrates a biologically important toxic endpoint (EPA 1985). For tests in which specific values were reported, EC_{50} values based on growth ranged from 138 µg/L to 1200 µg/L for diatoms *Isochrysis galbana* and *Skeletonema costatum*, respectively. The FPV for chlorpyrifos is 138 µg/L. None of the tests indicated that chlorpyrifos was more toxic to aquatic plants than to aquatic animals and criteria that protect aquatic animals will also protect aquatic plants.

HAZARD ASSESSMENT

Water Quality Criteria

The EPA (1985) guidelines specify using the lowest of three values, the FAV, the FCV, or the FPV, as a WQC (Appendix D). For chlorpyrifos, the lowest of these three values are the freshwater (0.02 µg/L) and saltwater (0.01 µg/L) FCVs, and these FCVs are proposed as the WQC. The freshwater WQC is recommended as interim because chronic toxicity data were lacking for a sensitive freshwater species. The WQC may be refined as more data become available.

The EPA acute and chronic freshwater WQC for chlorpyrifos are 0.083 µg/L and 0.041 µg/L, respectively, and the EPA acute and chronic saltwater WQC for chlorpyrifos are 0.011 µg/L and 0.0056 µg/L, respectively (EPA 1986) (Table 7). The EPA and CDFG have produced different numbers because additional toxicity data have been generated since development of the EPA criteria, and because CDFG used newer standards and guidelines to screen toxicity tests for acceptability. The data sets used in developing the EPA and the CDFG criteria may be compared by examining Table 1 of the EPA (1986) hazard assessment and Table A-1 of this hazard assessment. The CDFG and EPA criteria are similar and are approximately within a factor of two, a reasonable range considering test repeatability. Repeatability of acute tests generally is within a factor of two because CVs for most species vary from 30% to 50% (Peltier and Weber 1985).

Table 7. Freshwater and saltwater acute and chronic criteria for chlorpyrifos developed by U.S. Environmental Protection Agency (EPA) and California Department of Fish and Game (CDFG).

Criteria Type	EPA (µg/L)	CDFG (µg/L)	Ratio (EPA/DFG)
Freshwater Acute	0.083	0.07	1.2
Saltwater Acute	0.011	0.03	0.4
Freshwater Chronic	0.041	0.02	2.0
Saltwater Chronic	0.0056	0.01	0.6

The WQC proposed in this assessment are based on the toxicity of chlorpyrifos alone. Reevaluation of the WQC may be necessary if additive toxicity is demonstrated to occur when chlorpyrifos is combined with other pesticides commonly found in the Sacramento-San Joaquin River system such as diazinon, methyl parathion, and carbofuran. Data on the additive toxicity of chlorpyrifos with these other pesticides are not currently available, however other insecticides have demonstrated additive toxicity and it appears likely that chlorpyrifos toxicity would also be additive.

Hazard to Aquatic Animals

Chlorpyrifos has been detected in the San Joaquin River system at a peak concentration of 1.6 µg/L (Table 1). However, this concentration was detected only once; more typical concentrations ranged from 0.01 to 0.7 µg/L. Concentrations greater than the WQC of 0.02 µg/L have been detected in the San Joaquin River system for periods of time long enough to result in chronic exposure for aquatic organisms. Chlorpyrifos monitoring data were not available for saltwater.

A comparison of detected concentrations of chlorpyrifos (Table 1) with toxicity data and the interim freshwater WQC of 0.02 µg/L indicates that chlorpyrifos may pose a hazard to sensitive aquatic invertebrates in the Sacramento-San Joaquin River system.

The lowest 96-h LC_{50} value for freshwater invertebrates was 0.10 µg/L (mean value) for the cladoceran *Ceriodaphnia dubia*. No MATC values were available for freshwater invertebrates, however, the MATC value for the saltwater mysid *Mysidopsis bahia* is 0.003 µg/L.

Generally, invertebrates are more sensitive to chlorpyrifos than are fish. The lowest 96-h LC_{50} value for freshwater fish was 3.03 µg/L for bluegill. The only species of freshwater fish for which MATC values were available was the fathead minnow; the lowest MATC value for fathead minnow was 2.26 µg/L. Hazards to early life stages of fish may exist more frequently than hazards to adult fish.

Data Requirements

Acute toxicity data were available for all of the eight freshwater and saltwater families recommended by EPA (1985). Only one invertebrate species, the mysid *M. bahia*, has been tested under chronic exposure. Chronic toxicity data for freshwater invertebrates, particularly *Ceriodaphnia dubia*, are necessary to better define the FACR value. Additional chronic toxicity data for freshwater and saltwater species for which acute data are available would also be useful.

Sacramento-San Joaquin Estuary monitoring should be continued to help assess hazards posed by chlorpyrifos to aquatic species. Monitoring could be improved by lowering detection limits and by long-term continuous monitoring to define the environmental exposure to resident organisms.

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APPENDIX A. Abstracts of acute toxicity tests.

Accepted acute toxicity tests - The following tests used accepted test methods.

Borthwick and Walsh (1981) - In 1981, 96-h static toxicity tests were performed by the U.S. EPA on technical grade Dursban^R (97.7%) with juvenile mysid *Mysidopsis bahia* and 28-d old fry sheepshead minnow *Cyprinodon variegatus*. As part of this series of tests, 48-h static toxicity tests were performed with <2-h old eastern oyster *Crassostrea virginica*. ASTM (1978) testing guidelines were followed. Five concentrations of Dursban^R and solvent and dilution water controls were tested with mysids and sheepshead minnows. Seven concentrations of Dursban^R were tested with eastern oysters. Two replicates per concentration were tested with mysids and sheepshead minnows. Four replicates per concentration were tested with eastern oysters. Measurement of chlorpyrifos concentrations was not mentioned for any test. Water quality parameters during the eastern oyster, mysid, and sheepshead minnow tests averaged: temperature of 25.0 $\pm$1°C; Ph was not mentioned; dissolved oxygen level was not measured; and salinity of 20°/$_{oo}$. Control survival was greater than 90% for all tests. The 96-h LC_{50} values for the mysid and the sheepshead minnow were 0.056 µg/L and 270 µg/L, respectively. The 96-h EC_{50} value, based on abnormal development, for the eastern oyster was 1991 µg/L. Tests were also conducted using diatoms *Skeletonema costatum*, *Isochrysis galbana*, and *Thalassiosira pseudonana* (Appendix C).

Borthwick et al. (1985) - In 1985, 96-h flow-through toxicity tests were performed by the U.S. EPA on technical grade chlorpyrifos (92%) with 0-d, 7-d, 14-d, and 28-d old California grunion *Leuresthes tenuis*, Atlantic silverside *Menidia menidia*, and tidewater silverside *Menidia peninsulae*. ASTM (1980) testing guidelines were followed. Five concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. The number of replicates tested was not mentioned. Concentrations were measured. Water quality parameters during the test averaged: temperature of 25°C; pH was not mentioned; dissolved oxygen level was not mentioned; and salinity of 20°/$_{oo}$ in the *M. menidia* and *M. peninsulae* tests and 25°/$_{oo}$ in the *L. tenuis* tests. Control survival was not mentioned. The 96-h LC_{50} values for the 0-d, 7-d, 14-d, and 28-d old grunion were 1.0 µg/L, 1.0 µg/L, 1.0 µg/L and 1.3 µg/L, respectively. The 96-h LC_{50} values for the 0-d, 7-d, 14-d and 28-d old Atlantic silverside were 0.5 µg/L, 1.0 µg/L, 1.1 µg/L, and 3.0 µg/L, respectively. The 96-h LC_{50} values for the 0-d, 7-d, 14-d and 28-d old tidewater silversides were 1.0 µg/L, 0.5 µg/L, 0.4 µg/L, 0.9 µg/L, respectively.

<u>California Department of Fish and Game (CDFG) (1992a)</u> - In 1992, 96-h static renewal toxicity tests (Test No. 92-133) were performed on technical grade chlorpyrifos (99%) with neonate mysid *Neomysis mercedis*. ASTM (1992) testing guidelines were followed. Five concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. Two replicates per concentration were tested. Chlorpyrifos concentrations were measured at the beginning and end of each test and averaged 86 to 124% of nominal concentrations. Water quality parameters during the test averaged: temperature of 17.2°C; pH of 8.4; dissolved oxygen level of 8.4 mg/L; hardness of 499.0 mg/L; conductivity of 3096 µs/cm; and alkalinity of 154.0 mg/L. Control survival was 95% in the solvent control and 100% in the dilution water control. The 96-h LC_{50} value for *N. mercedis* was 0.16 µg/L.

<u>CDFG (1992a)</u> - In 1992, 96-h static renewal toxicity tests (Test No. 92-142) were performed on technical grade chlorpyrifos (99%) with neonate mysid *Neomysis mercedis*. ASTM (1992) testing guidelines were followed. Five concentrations of chlorpyrifos

were tested and solvent and dilution water controls were used. Two replicates per concentration were tested. Chlorpyrifos concentrations were measured at the beginning and end of each test and averaged 71 to 84% of nominal concentrations. Water quality parameters during the test averaged: temperature of 17.1°C; pH of 8.4; dissolved oxygen level of 9.3 mg/L; hardness of 509.0 mg/L; conductivity of 3151 µs/cm; and alkalinity of 151.0 mg/L. Control survival was 100% for both solvent and dilution water controls. The 96-h LC_{50} value for *N. mercedis* was 0.14 µg/L.

CDFG (1992a) - In 1992, 96-h static renewal toxicity tests (Test No. 92-143) were performed by CDFG on technical grade chlorpyrifos (99%) with neonate mysid *Neomysis mercedis*. ASTM (1992) testing guidelines were followed. Five concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. Two replicates per concentration were tested. Chlorpyrifos concentrations were measured at the beginning and end of each test and averaged 71 to 84% of nominal concentrations. Water quality parameters during the test averaged: temperature of 17.4°C; pH of 8.2; dissolved oxygen level of 8.9 mg/L; hardness of 515.0 mg/L; conductivity of 3192 µs/cm; and alkalinity of 151.5 mg/L. Control survival was 100% for both solvent and dilution water controls. The 96-h LC_{50} value for *N. mercedis* was 0.15 µg/L.

CDFG (1992b) - In 1992, 96-h static toxicity tests (Test No. 92-139) were performed by the CDFG on technical grade chlorpyrifos (99%) with neonate cladoceran *Ceriodaphnia dubia*. EPA (1989) and ASTM (1988a,b) testing guidelines were followed. Five concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. Nine replicates per concentration were tested. Chlorpyrifos concentrations were measured at the beginning and end of each test and averaged 75 to

100% of nominal concentrations. Water quality parameters during the test averaged: temperature of 24.3°C, pH of 8.2; dissolved oxygen level of 7.7 mg/L; hardness of 121.5 mg/L; conductivity of 333.7 µs/cm; and alkalinity of 105.0 mg/L. Control survival was 90% in both the solvent and dilution water controls. The 96-h LC_{50} value for *C. dubia* was 0.08 µg/L.

CDFG (1992b) - In 1992, 96-h static toxicity tests (Test No. 92-150) were performed by the CDFG on technical grade chlorpyrifos (99%) with neonate cladoceran *Ceriodaphnia dubia*. EPA (1989) and ASTM (1988a,b) testing guidelines were followed. Five concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. Nine replicates per concentration were tested. Chlorpyrifos concentrations were measured at the beginning and end of each test and averaged 94 to 105% of nominal concentrations. Water quality parameters during the test averaged: temperature of 24.6°C; pH of 8.3; dissolved oxygen level of 7.7 mg/L; hardness of 120.0 mg/L; conductivity of 325.7 µs/cm; and alkalinity of 107.0 mg/L. Control survival was 100% in both solvent and dilution water controls. The 96-h LC_{50} value for *C. dubia* was 0.13 µg/L.

Clark et al (1985) - In 1985, 96-h static and flow-through toxicity tests were performed by the U.S. EPA on technical grade chlorpyrifos (percent active ingredient not specified) with tidewater silverside *Menidia peninsulae* and inland silverside *Menidia beryllina*. ASTM (1980) testing guidelines were followed. Five concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. One replicate per concentration was tested. Chlorpyrifos concentrations were measured at 48-h and 96-h but values were not mentioned. Water temperatures in the tidewater and inland silverside tests averaged 24.6°C and 24.5°C, respectively. Other water quality parameters during the test were measured but values were not given. Solvent and dilution water controls were used and survival was within ASTM guidelines. The 96-h LC_{50} values for tidewater silverside and inland silverside were 1.3 µg/L and 4.2 µg/L, respectively. Although this study had some deficiencies it was considered acceptable because of the reputation of the laboratory, ASTM guidelines were followed, and control survival was acceptable.

Federle and Collins (1976) - In 1976, 96-h static toxicity tests were performed by the Department of Entomology, Ohio State University on technical grade chlorpyrifos (94%) with adult crawling water beetle *Petodytes* sp. Commonly recognized testing guidelines were not mentioned. Four concentrations of chlorpyrifos were tested and a solvent control was used. Three replicates per concentration were tested. All concentrations tested were nominal. Water quality parameters during the test averaged: temperature of 25 $\pm$2°C; pH of 7.4; dissolved oxygen level was not mentioned but test solutions were aerated. Control survival was 95%. The 96-h LC_{50} value for *Petodytes* sp. was 0.8 µg/L.

Hansen et al. (1986) - In 1986, 96-h static toxicity tests were performed by the U.S. Environmental Protection Agency on technical grade chlorpyrifos (92%) with 2-mo. old gulf toadfish *Opsanus beta*. ASTM (1985) proposed testing guidelines were followed. Five concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. One replicate per concentration was tested. Chlorpyrifos concentrations averaged 50 to 60% of nominal concentrations. Water quality parameters during the test averaged: temperature of 25 to 27°C; pH was not mentioned; dissolved oxygen was not measured; and salinity of $29^{\circ}/_{\circ\circ}$ to $30^{\circ}/_{\circ\circ}$. Control survival was 100%. The 96-h LC_{50} value for *Opsanus beta* was 520 µg/L.

Holcombe et al. (1982) - In 1982, a 96-h flow-through toxicity test was performed by the U.S. EPA on technical grade Dursban[R] (99.9%) with juvenile rainbow trout *Oncorhynchus mykiss* and 31-d to 32-d old fathead minnow *Pimephales promelas*. APHA (1975) and EPA (1975) testing guidelines were followed. Five concentrations of Dursban[R] and a water control were tested in each test. Two replicates per concentration were tested. Chlorpyrifos concentrations were measured daily and averaged 88 to 112% of nominal concentrations in rainbow trout tests and 84 to 116% in fathead minnow tests. Water quality parameters during the test averaged: temperature of 15.6 $\pm$1.8°C for rainbow trout tests and 25.1 $\pm$1.3°C for fathead minnow tests; pH of 7.0 to 7.4; dissolved oxygen level of 9.3 mg/L for rainbow trout tests and 7.3 mg/L for fathead minnow tests; hardness of 45.3 mg/L; and alkalinity of 41.8 mg/L. Control survival was 100% in both tests. The 96-h LC_{50} value for the fathead minnow was 203 µg/L. The 96-h LC_{50} value for the rainbow trout was 8.0 µg/L.

Jarvinen and Tanner (1982) - In 1982, 96-h flow-through toxicity tests were performed by the U.S. EPA on technical grade chlorpyrifos (98.7%) with larval fathead minnow *Pimephales promelas*. APHA (1975) testing guidelines were followed. Five concentrations of chlorpyrifos were tested and a dilution water control was used. Two replicates per concentration were tested. Measured concentrations averaged 102 to 144% of nominal concentrations. Water quality parameters during the test averaged: temperature of 25.0 $\pm$0.6°C; pH of 7.4 to 7.8; dissolved oxygen level of 6.5 to 8.4 mg/L; hardness of 45.8 mg/L; and alkalinity of 43.1 mg/L. Control survival was 100%. The 96-h LC_{50} value for *P. promelas* was 140 µg/L.

Kersting and Van Wijngaarden (1992) - In 1992, a 48-h static toxicity test was performed by the Research Institute for Nature Management in The Netherlands on technical grade chlorpyrifos

(99%) with <24-h old cladoceran *Daphnia magna*. Commonly recognized test guidelines were not mentioned. Six concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. Two replicates per concentration were tested. Chlorpyrifos concentrations were measured at the beginning of each test and averaged 57% of nominal concentrations. Water quality parameters during the test averaged: temperature of 19.5 $\pm$0.5°C; pH of 6.8 to 7.0; and dissolved oxygen level of 7.7 mg/L to 8.8 mg/L. Control survival was 100%. The 48-h LC_{50} value for *D. magna* was 1.0 µg/L.

Macek et al. (1969) - In 1969, 96-h static toxicity tests were performed by the U.S. Bureau of Sport Fisheries and Wildlife on technical grade Dursban[R] (97%) with rainbow trout *Oncorhynchus mykiss*. Commonly recognized testing guidelines were not mentioned. Seven concentrations of chlorpyrifos and a solvent control were tested. One replicate per concentration was tested. All concentrations were nominal. Water quality parameters during the test averaged: temperature of 1.6 $\pm$0.6°C in one series of tests, 7.2 $\pm$0.6°C in a second series of tests, and 12.7 $\pm$0.6°C in a third series of tests; pH of 7.1; dissolved oxygen level was not measured but solutions used in tests were well aerated; and alkalinity of 35 mg/L. Control survival was 100%. The 96-h LC_{50} value for *O. mykiss* at 1.6°C was 51 µg/L. The 96-h LC_{50} value for *O. mykiss* at 7.2°C was 15 µg/L. The 96-h LC_{50} values for *O. mykiss* at 12.7°C was 7.1 µg/L. Only the test performed at 12.7 $\pm$0.6° C was used because it most closely adhered to ASTM (1988a) standards for trout tests.

Mayer (1987) - In 1987, results of acute toxicity tests on 197 chemicals with 52 estuarine and marine species were compiled. All tests were performed at the Environmental Research Laboratory, Gulf Breeze, Florida during 1961 to 1986. The tests used technical grade chlorpyrifos (92%) and generally complied with ASTM (1980) standards. At least four concentrations of chlorpyrifos were tested in each test. Depending on the species, temperatures ranged from 11°C to 31°C. Dissolved oxygen, pH, control survival, water hardness, and chlorpyrifos concentrations were not given. The 96-h and 48-h LC_{50} values and the 48-h EC_{50} values are listed in Table A-1. Although information about some important test characteristics could not be obtained, most of these data were accepted because of the use of ASTM guidelines and the reputation of the laboratory. Acceptable data were available for Atlantic and tidewater silversides, blue crab, brown, grass, and pink shrimps, California grunion, gulf and longnose killifish, gulf toadfish, *Mysidopsis bahia*, sheepshead minnow, and striped mullet.

<u>Mayer and Ellersieck (1986)</u> - In 1986, a study was conducted by the Fish and Wildlife Service to generate static acute toxicity test data for 410 chemicals with 66 freshwater species. All tests were performed at the Columbia National Fisheries Research Laboratory and its field laboratories between 1965 to 1984. The studies on technical grade chlorpyrifos (97%) were conducted with eight species. The tests were generally in compliance with ASTM (1980) and EPA (1975) standards. At least five concentrations of chlorpyrifos were tested. Two replicates per concentration were tested. Depending on the species, water quality parameters during the tests were as follows: temperature of 2.0°C to 29°C; pH of 6.0 to 9.0; and hardness of 44 mg/L to 272 mg/L. Control survival, dissolved oxygen, and measurement of chlorpyrifos concentrations were not discussed. The 96-h LC_{50} values are listed in Table A-1. Although information about some important test characteristics could not be obtained, most of these data were accepted because of the use of ASTM guidelines and the reputation of the laboratory. Acceptable data were available for bluegill, channel catfish, and lake and cutthroat trouts.

Phipps and Holcombe (1985) - In 1985, 96-h flow-through toxicity tests were performed by the U.S. EPA on technical grade chlorpyrifos (99.9%) with rainbow trout *Oncorhynchus mykiss*, fathead minnow *Pimephales promelas*, goldfish *Carassius auratus*, channel catfish *Ictalurus punctatus*, bluegill *Lepomis macrochirus*, crayfish *Orconectes immunis,* and snail *Aplexa hypnorum*. ASTM (1980) and APHA (1980) testing guidelines were followed. Three or five concentrations of chlorpyrifos were tested and water controls were used. One replicate per concentration was tested in all tests. Chlorpyrifos concentrations were measured daily and averaged 85 to 114% of nominal concentrations. Water quality parameters during the test averaged: temperature of 17.3 $\pm$0.6°C; pH of 7.1 to 7.8; dissolved oxygen level of 7.5 $\pm$1.6 mg/L; hardness of 44.4 mg/L; and alkalinity of 45.4 mg/L. Control survival was 100%. The 96-h LC_{50} value for rainbow trout, fathead minnow, goldfish, channel catfish, bluegill, crayfish, and snails were 9 µg/L, 542 µg/L, >806 µg/L, 806 µg/L, 10 µg/L, 6.0 µg/L, and >806 µg/L, respectively.

Sanders (1969) - In 1969, 96-h static toxicity tests were performed by the U.S. Bureau of Sport Fisheries and Wildlife on technical grade Dursban[R] (97%) with 2 month old ($\pm$5 days) amphipod *Gammarus lacustris*. Commonly recognized testing guidelines were not mentioned. Five concentrations of Dursban[R] were tested and a dilution water control was used. One replicate per concentration was tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 70 $\pm$1°F; pH of 7.1; dissolved oxygen was not measured but test water was aerated for 10 minutes before testing began; and alkalinity of 30.0 mg/L. Control survival was 100%. The 96-h LC_{50} value for *G. lacustris* was 0.11 µg/L.

Sanders and Cope (1968) - In 1968, 96-h static toxicity tests were performed by the U.S. Bureau of Sport Fisheries and Wildlife on technical grade Dursban[R] (97%) with naiad stoneflies *Pteronarcys californica*, *Pteronarcella badia*, and *Claassenia sabulosa*. Commonly recognized testing guidelines were not mentioned. Four concentrations of Dursban[R] were tested and a dilution water control was used. One replicate per concentration was tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 15.5 $\pm$0.5°C; pH of 7.1; dissolved oxygen level of 7 mg/L initially, 5 mg/L after 24-h, and 3 mg/L at 96-h; alkalinity of 35 mg/L. Control survival was 100%. The 96-h LC_{50} values for *Pteronarcys californica*, *Pteronarcella badia*, and *Claassenia sabulosa* were 10 µg/L, 0.38 µg/L, and 0.57 µg/L, respectively. Although the dissolved oxygen was too low, these tests were accepted because control survival was 100%

Unaccepted acute toxicity tests - The following tests did not use accepted test methods and/or produce accepted results.

Acevedo (1991) - In 1991, 96-h static and flow-through toxicity tests were performed by the Hawaii Institute of Marine Biology on technical grade chlorpyrifos (percent active ingredient not specified) with coral planulae *Pocillopora damicornis*. EPA (1988) testing guidelines were followed. Four concentrations of chlorpyrifos were tested. Three replicates per concentration were tested. The use of controls was not mentioned. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the study averaged: temperature of 25-27°C; pH was not mentioned; dissolved oxygen level was not mentioned; and salinity was not mentioned. Control survival was not mentioned. The static and flow-through LC_{50} values were not reported. This test was not used because essential information, such as toxicity values, and control survival, was lacking.

Ali and Majori (1982) - In 1982, 24-h toxicity tests were performed by the University of Florida on technical grade chlorpyrifos (percent active ingredient not specified) with fourth instar larvae of midge *Chironomus salinarius*. Commonly recognized testing guidelines were not mentioned. Four to five concentrations and a dilution water control were tested. Three replicates per concentration were tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 27 $\pm$2°C; pH of 7.8 to 9.0; dissolved oxygen level of <2 to 10 mg/L; and salinity of 27 to 45°/∞. Control survival was not mentioned. The 24-h LC_{50} value for *C. salinarius* was 0.44 µg/L. This value was not used because the organisms had been exposed to pesticides prior to testing, dissolved oxygen levels fell below an acceptable range, and essential information, such as control survival, was lacking.

CDFG (1992b) - In 1992, 96-h static toxicity tests (Test No. 92-137) were performed by CDFG on technical grade chlorpyrifos (99%) with neonate cladoceran *Ceriodaphnia dubia*. EPA (1989) and ASTM (1992a,b) testing guidelines were followed. Five concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. Nine replicates per concentration were tested. Chlorpyrifos concentrations were measured at the beginning and end of each test and averaged 105 to 150% of nominal concentrations. Water quality parameters during the test averaged: temperature of 24.5°C; pH of 8.4; dissolved oxygen level of 7.7 mg/L; hardness of 119.0 mg/L; conductivity of 345.6 µs/cm; and alkalinity of 105.5 mg/L. Control survival was 70% in the solvent control and 100% in the dilution water control. The 96-h LC_{50} value for *C. dubia* was 0.12 µg/L. This value was not used because solvent control survival was less than 90%.

Carter and Graves (1973) - In 1973, 96-h static toxicity tests were performed by the Department of Entomology, Louisiana State University on chlorpyrifos (percent active ingredient not specified) with White River crayfish *Procambarus acutus*, bluegill *Lepomis macrochirus*, mosquitofish *Gambusia affinis*, and channel catfish *Ictalurus punctatus*. APHA (n.d.) testing guidelines were followed. The number of chlorpyrifos concentrations tested and use of controls were not mentioned. Five replicates per concentration were used in the crayfish tests. Two replicates per concentration were used in all other tests. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 26°C in crayfish and channel catfish tests, 23°C in bluegill tests, and 24°C in mosquitofish tests; pH was not mentioned for any test; and dissolved oxygen level of 7 mg/L to 10 mg/L in bluegill and channel catfish tests, 9 mg/L to 11 mg/L in White River crawfish tests, and 9 mg/L in mosquitofish tests. The 96-h LC_{50} values for crayfish, bluegill, mosquitofish, and channel catfish were 2 µg/L, 30 µg/L, 280 µg/L, and 160 µg/L, respectively. These values were not used because essential information, such as control survival and concentrations tested, was lacking.

Cebrian et al. (1992) - In 1992, 96-h static toxicity tests were performed by the Department of Animal Biology, University of Valencia, Spain on technical grade chlorpyrifos (99.8%) with crayfish *Procambarus clarkii*. EPA (1975) testing guidelines were followed. The chlorpyrifos concentrations and number of replicates tested were not mentioned. A solvent control was used. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 22 $\pm$1°C; pH of 7.9 $\pm$0.2; dissolved oxygen level was not mentioned; hardness of 250 mg/L as $CaCO_3$; and alkalinity of 4.1 mM/L. Control survival was not mentioned. The 96-h LC_{50} value for *P. clarkii* was 21 µg/L, respectively. This value was not used because essential information, such as control survival and concentrations tested, was lacking.

Darwazeh and Mulla (1974) - In 1974, 92-h toxicity tests were performed by the Department of Entomology, University of California at Riverside on technical chlorpyrifos (percent active ingredient not specified) with mosquitofish *Gambusia affinis*. Commonly recognized testing guidelines were not mentioned. The chlorpyrifos concentrations tested were not mentioned. Two replicates per concentration were tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test were not mentioned. Control survival was not mentioned. LC_{50} values were not determined in this study. The 92-h LC_{85} value for *G. affinis* was 1000 µg/L. The test was unacceptable because essential information, such as water quality parameters and concentrations tested, was lacking, the test duration was less than 96-h, and LC_{50} values were not determined.

Davey et al. (1976) - In 1976, 72-h static toxicity tests were performed by the Department of Entomology, University of Arkansas on chlorpyrifos (percent active ingredient not specified) with mature mosquitofish *Gambusia affinis* and green sunfish *Lepomis cyanellus*. Commonly recognized testing guidelines were not mentioned. One concentration of chlorpyrifos was tested and a solvent control was used. Two replicates per concentration were tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test were not mentioned. Control survival was not mentioned. The 72-h LC_{50} values for the mosquitofish and the green sunfish were 260 µg/L and 40 µg/L, respectively. These values were not used because the test duration was too short, an inadequate number of concentrations was tested, and essential information, such as water quality parameters and control survival, was lacking.

Earnest (1970) - In 1970, 96-h static and flow-through toxicity tests were performed by the U.S. Bureau of Sport Fisheries and Wildlife on technical grade Dursban® (99%) with Korean shrimp *Palaemon macrodactylus*. A 96-h flow-through toxicity test was performed on Dursban® (90%) with striped bass *Morone saxatilis*. Commonly recognized testing guidelines were not mentioned. Dursban® concentrations, number of replicates, and use of controls were not mentioned. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 12.2°C to 12.8°C; pH was not mentioned; dissolved oxygen level was not mentioned; and salinity of 15‰ to 30‰. Control survival was not mentioned. The 96-h static and flow-through TL_{50}s for the Korean shrimp were 0.25 µg/L and 0.01 µg/L, respectively. The 96-h flow-through TL_{50} for the striped bass was 0.58 µg/L. These values were not used because essential information, such as control survival and use of controls, was lacking.

El-Refai et al. (1976) - In 1976, 48-h static toxicity tests were performed by Al-Azhar University Cairo on Dursban® (40.8%) with fingerling tilapia *Tilapia nilotica* and carp *Cyprinus carpio*. Commonly recognized testing guidelines were not mentioned. One replicate per concentration was tested when larger fish were used, otherwise replicates were not used. Chlorpyrifos concentrations were measured but values were not given. Water quality parameters during the test averaged: temperature of 22°C to 25°C; pH of 7.8 to 8.2; dissolved oxygen level of 6.8 mg/L to 7.4 mg/L; hardness of 116 mg/L to 123 mg/L; conductivity of 270 to 300 µmho/cm; and alkalinity of 122 mg/L to 125 mg/L. Control survival was not mentioned. For smaller carp, the 48-h LC_{50} value was 280 µg/L. For larger carp, the 48-h LC_{50} value was 59 µg/L. For smaller tilapia, the 48-h LC_{50} value was 62 µg/L. For larger tilapia, the 48-h LC_{50} value was 114 µg/L. These values were not used because essential information, such as control survival, was lacking, the test duration was less than 96-h, the pesticide formulation used was too low in active ingredient, and the temperature varied by more than 2°C.

Ferguson et al. (1966) - In 1966, 36-h toxicity tests were performed by the Department of Zoology, Mississippi State University on technical grade chlorpyrifos (99 ±1%) with the golden shiner *Notemigonus crysoleucas*, mosquitofish *Gambusia affinis*, and green sunfish *Lepomis cyanellus*. Commonly recognized testing guidelines were not mentioned. Chlorpyrifos concentrations and number of replicates tested were not mentioned. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature was not mentioned; pH of 7.4; dissolved oxygen level was not mentioned; and hardness of 24 mg/L. Control survival was 95%. The 36-h LC_{50} values for the golden shiner, mosquitofish, and green sunfish were 35 µg/L to 125 µg/L, 215 µg/L to 595 µg/L, and 22.5 µg/L to 125 µg/L, respectively. These values were not used because test duration was less than 96-h and organisms were exposed to pesticides prior to testing.

Ferrando et al. (1991) - In 1991, 96-h flow-through toxicity tests were performed by the Department of Animal Biology, University of Valencia, Spain on technical grade chlorpyrifos (97%) with the European eel *Anguilla anguilla*. U.S. EPA (1975) testing guidelines were followed. Chlorpyrifos concentrations tested were not mentioned. Solvent and dilution water controls were used. Three replicates per concentration were tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 20°C; pH of 7.9 ±0.2; dissolved oxygen level was not mentioned;

alkalinity of 4.1 mmol/L; and a hardness of 250 mg/L. Control survival was 100%. The 96-h LC_{50} value for *A. anguilla* was 540 µg/L. This value was not used because dissolved oxygen was not measured and essential information, such as concentrations tested, was lacking.

Ferrando and Andreu-Moliner (1991) - In 1991, 24-h static toxicity tests were performed by the Department of Animal Biology, University of Valencia, Spain on chlorpyrifos (percent active ingredient not specified) with newly hatched rotifera: *Brachionus calyciflorus* and *Brachionus plicatilis*. EPA (1985) testing guidelines were followed. Five concentrations of chlorpyrifos were tested. Use of a control was not mentioned. Three replicates per concentration were tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 25°C; pH of 7.4 to 7.8 in *B. calyciflorus* tests and 7.7 in *B. plicatilis* tests; hardness of 80 mg/L to 100 mg/L in *B. calyciflorus* tests (hardness not mentioned in *B. plicatilis* tests); and salinity of $15^{\circ}/_{\circ\circ}$ in *B. plicatilis* tests (salinity not mentioned in B. calyciflorus tests). Control survival was 100% for both *B. calyciflorus* and *B. plicatilis* tests. The 24-h LC_{50} value for *B. calyciflorus* and *B. plicatilis* were 11850 µg/L and 10670 µg/L, respectively. These values were not used because essential information, such as percent active ingredient, was lacking.

Holbrook (1982) - In 1982, 24-h static toxicity tests were performed by the U.S. Department of Agriculture on technical grade chlorpyrifos (percent active ingredient not specified) with fourth instar larvae of ceratopogonid, *Culicoides variipennis*. Commonly recognized testing guidelines were not mentioned. Four concentrations of chlorpyrifos were tested and a dilution water control was used. Two replicates per concentration were tested. Measurement of chlorpyrifos concentrations was not mentioned.

Water quality parameters during the test averaged: temperature of 22 ±1°C; pH of 7.0; and dissolved oxygen was not measured. Control survival was not mentioned. The 24-h LC_{50} value for *C. variipennis* was 4.3 µg/L. This value was not used because test duration should have been 96-h (ASTM 1988a) and information for several test parameters were missing.

<u>Holbrook (1983)</u> - In 1983, 24-h static toxicity tests were performed by the U.S. Department of Agriculture on technical grade chlorpyrifos (percent active ingredient not specified) with larvae of ceratopogonid, *Culicoides variipennis*. Commonly recognized testing guidelines were not mentioned. Five concentrations of chlorpyrifos were tested and a dilution water control was used. Three replicates per concentration were tested. All concentrations were nominal. Water quality parameters during the test averaged: temperature of 22 ±1°C; neutral pH; dissolved oxygen was not measured but larvae remained close to the surface; and soft water was used. Control survival was greater than 90%. The 24-h LC_{50} value for *C. variipennis* was 2.96 µg/L. This value was not used because test duration should have been 96-h (ASTM 1988a) and information for several test parameters were missing.

<u>Kenaga et al. (1965)</u> - In 1965, 24-h toxicity tests were performed by the Dow Chemical Company on Dursban® (percent active ingredient not specified) with adult and nymph cladoceran *Daphnia sp.*, immature goldfish *Carrassius auratus*, and mature snail *Helisoma trivolvis*. Chemical Specialties Manufacturers Association (1963) testing guidelines were followed. Three concentrations of Dursban® were tested. There was no mention of the use of controls. The number of replicates tested was not mentioned. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 65°F; pH was not mentioned; dissolved oxygen level

was not mentioned; and hardness was not mentioned. Control survival was not mentioned. The 24-h LC_{50} value for cladoceran, goldfish, and snail were 16 µg/L, 180 µg/L, and >2000 µg/L, respectively. These values were not used because acute toxicity tests using fish must be 96-h in duration (ASTM 1988a), and essential information, such as the use of controls and control survival, was lacking.

Sanders (1972) - In 1972, 96-h static toxicity tests were performed by the U.S. Bureau of Sport Fisheries and Wildlife on technical grade Dursban^R (97%) with amphipod *Gammarus fasciatus*. Commonly recognized testing guidelines were not mentioned. Five concentrations of Dursban^R were tested. The use of a control was not mentioned. One replicate per concentration was tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 21 ±0.5°C; pH of 7.1; dissolved oxygen level of 8 mg/L; and alkalinity of 35 mg/L. Control survival was not mentioned. The 96-h LC_{50} value for *G. fasciatus* was 0.32 µg/L. This value was not used because essential information, such as the use of controls and control survival, was lacking.

Schimmel et al. (1983) - In 1983, 96-h flow-through toxicity tests were performed by the U.S. EPA on technical grade chlorpyrifos (92%) with Atlantic silverside *Menidia menidia*, mysid *Mysidopsis bahia*, sheepshead minnow *Cyprinodon variegatus*, longnose killifish *Fundulus similis*, and striped mullet *Mugil cephalus*. ASTM (1980) testing guidelines were followed. Chlorpyrifos concentrations tested were not mentioned. Solvent and dilution water controls were used. Three replicates per concentration were tested in mysid tests. The number of replicates tested in fish tests was not mentioned. Chlorpyrifos concentrations were measured but values were not given. Water quality parameters during the test were not mentioned. Control survival was not mentioned. The 96-h LC_{50} value for the Atlantic silverside, mysid, sheepshead minnow, longnose killifish, and striped mullet were 1.7 µg/L, 0.035 µg/L, 136 µg/L, 4.1 µg/L, and 5.4 µg/L, respectively. These values were not used because essential information, such as water quality parameters and control survival, was lacking.

<u>Strickman (1985)</u> - In 1985, 7-d static toxicity tests were performed by the US Air Force Occupational and Environmental Health Laboratory on technical grade chlorpyrifos (93 to 100%) with second instar mosquito *Wyeomyia smithii*. Commonly recognized testing guidelines were not mentioned. Three concentrations of chlorpyrifos were tested and a solvent control was used. Eight replicates per concentration were tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 27°C; pH was not mentioned; dissolved oxygen level was not mentioned; hardness was not mentioned. Control survival was not mentioned. The data from this study were not used because essential information, such as LC_{50} value, control survival, and dissolved oxygen level, was lacking.

<u>Thirugnanum and Forgash (1977)</u> - In 1977, 96-h flow-through toxicity tests were performed by the Department of Entomology and Economic Zoology, New Brunswick State University on technical grade chlorpyrifos (99.5%) with mummichog *Fundulus heteroclitus*. Commonly recognized testing guidelines were not mentioned. Five concentrations of chlorpyrifos were tested and a solvent control was used. Two replicates per concentration were tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 25°C; pH of 7.5 to 8.0; dissolved oxygen level was not mentioned but the testing solution was aerated; and salinity of 20°/$_{oo}$ to 25°/$_{oo}$. Control survival was not mentioned. The 96-h LC_{50} value

for *F. heteroclitis* was 4.65 µg/L. The 96-h LC_{50} value for *F. heteroclitis* was not used because essential information, such as control survival, was lacking.

U.S. Army Environmental Hygiene Agency (1970) - In 1970, toxicity tests were performed by the U.S. Army Environmental Hygiene Agency on Dursban[R] (percent active ingredient not specified) with first instar larvae of mosquito *Culicoides pipiens quinquefasciatus*. Commonly recognized testing guidelines were not mentioned. Eight concentrations of Dursban[R] were tested and a solvent control was used. Four replicates per concentration were tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test were not mentioned. Control survival was not mentioned. The test found that sublethal concentrations of Dursban[R] had no effect on larval development of *C. pipiens quinquefasciatus*. This information was not used because essential information, such as acute toxicity data and control survival, was lacking.

Walton et al. (1990) - In 1990, 24-h static toxicity tests were performed by the Department of Entomology, University of California at Riverside on chlorpyrifos (the percent active ingredient not specified) with 4-5 day old tadpole shrimp *Triops longicaudatus*. Commonly recognized testing guidelines were not mentioned. Measurement of chlorpyrifos concentrations was not mentioned. A solvent control was used. Ten replicates per concentration were tested. Water quality parameters during the test averaged: temperature of 28 ± 2°C; pH was not mentioned; dissolved oxygen level was not mentioned. Control survival was not mentioned. The 24-h LC_{50} value for *T. longicaudatus* was 4.0 µg/L. This value was not used because essential information, such as concentrations tested and control survival, was lacking.

Table A-1. Values (μg/L) from accepted tests on the acute toxicity of chlorpyrifos to aquatic animals.

Species	Life Stage[a]	Method[b]	Formulation	Salinity/ Hardness[a]	Test Length	Effect	Values (95% C.L.)[c]	Reference
Amphipod *Gammarus lacustris*	2 mo.	S,U	Tech(97%)	N/A	96-h	LC_{50}	0.11 (0.071-0.17)	Sanders 1969
Atlantic silverside *Menidia menidia*	juv. 53-d	F,M	Tech(92%)	24°/oo	96-h	LC_{50}	1.7 (1.4-2.0)	Mayer 1987
Atlantic silverside *Menidia menidia*	0-d	F,M	Tech(92%)	20°/oo	96-h	LC_{50}	0.5 (0.4-0.7)	Borthwick et al. 1985
Atlantic silverside *Menidia menidia*	7-d	F,M	Tech(92%)	20°/oo	96-h	LC50	1.0 (0.9-1.2)	Borthwick et al. 1985
Atlantic silverside *Menidia menidia*	14-d	F,M	Tech(92%)	20°/oo	96-h	LC_{50}	1.1 (N/A)	Borthwick et al. 1985
Atlantic silverside *Menidia menidia*	28-d	F,M	Tech(92%)	20°/oo	96-h	LC_{50}	3.0 (2.6-4.0)	Borthwick et al. 1985
Blue crab *Callinectes sapidus*	juv.	F,U	Tech(92%)	20°/oo	48-h	EC_{50}	5.2 (N/A)	Mayer 1987
Bluegill *Lepomis macrochirus*	N/A	S,U	Tech(97%)	44 mg/L as $CaCO_3$	96-h	LC_{50}	2.4 (1.1-5.1)	Mayer and Ellersieck 1986
Bluegill *Lepomis macrochirus*	N/A	S,U	Tech(97%)	272 mg/L as $CaCO_3$	96-h	LC_{50}	4.2 (3.2-5.5)	Mayer and Ellersieck 1986
Bluegill *Lepomis macrochirus*	N/A	S,U	Tech(97%)	272 mg/L as $CaCO_3$	96-h	LC_{50}	1.8 (1.5-2.2)	Mayer and Ellersieck 1986
Bluegill *Lepomis macrochirus*	N/A	S,U	Tech(97%)	272 mg/L as $CaCO_3$	96-h	LC_{50}	2.5 (1.2-3.0)	Mayer and Ellersieck 1986
Bluegill *Lepomis macrochirus*	N/A	S,U	Tech(97%)	272 mg/L as $CaCO_3$	96-h	LC_{50}	1.7 (N/A)	Mayer and Ellersieck 1986

Table A-1. Continued -2-

Species	Life Stage[a]	Method[b]	Formulation	Salinity/ Hardness[a]	Test Length	Effect	Values (95% C.L.)[c]	Reference
Bluegill *Lepomis macrochirus*	N/A	F,M	Tech(99.9%)	44.4 mg/L as $CaCO_3$	96-h	LC_{50}	10 (6-14)	Phipps and Holcombe 1985
Brown shrimp *Penaeus aztecus*	juv.	F,U	Tech(92%)	26‰	48-h	EC_{50}	0.20 (N/A)	Mayer 1987
California grunion *Leuresthes tenuis*	0-d	F,M	Tech(92%)	25‰	96-h	LC_{50}	1.0 (0.8-1.3)	Borthwick et al. 1985
California grunion *Leuresthes tenuis*	7-d	F,M	Tech(92%)	25‰	96-h	LC_{50}	1.0 (0.8-1.3)	Borthwick et al. 1985
California grunion *Leuresthes tenuis*	14-d	F,M	Tech(92%)	25‰	96-h	LC_{50}	1.0 (0.8-1.4)	Borthwick et al. 1985
California grunion *Leuresthes tenuis*	28-d	F,M	Tech(92%)	25‰	96-h	LC_{50}	1.3 (1.0-1.7)	Borthwick et al. 1985
California grunion *Leuresthes tenuis*	larvae 7-d	F,M	Tech(92%)	25‰	96-h	LC_{50}	2.7 (1.9-5.4)	Mayer 1987
California grunion *Leuresthes tenuis*	larvae 14-d	F,M	Tech(92%)	20‰	96-h	LC_{50}	1.0 (0.76-1.4)	Mayer 1987
Channel catfish *Ictalurus punctatus*	N/A	F,M	Tech(99.9%)	44.4 mg/L as $CaCO_3$	96-h	LC_{50}	806 (434-1088)	Phipps and Holcombe 1985
Channel catfish *Ictalurus punctatus*	N/A	S,U	Tech(97%)	44 mg/L as $CaCO_3$	96-h	LC_{50}	280 (206-381)	Mayer and Ellersieck 1986
Cladoceran *Ceriodaphnia dubia*	neonate <24-h	S,M	Tech(99%)	120 mg/L as $CaCO_3$	96-h	LC_{50}	0.13 (0.08-0.16)	CDFG 1992b
Cladoceran *Ceriodaphnia dubia*	neonate <24-h	S,M	Tech(99%)	122 mg/L as $CaCO_3$	96-h	LC_{50}	0.08 (0.06-0.11)	CDFG 1992b

Table A-1. Continued -3-

Species	Life Stage[a]	Method[b]	Formulation	Salinity/ Hardness[a]	Test Length	Effect	Values (95% C.L.)[c]	Reference
Cladoceran *Daphnia magna*	neonate <24-h	S,M	Tech(99%)	N/A	48-h	LC_{50}	1.0 (1.0-1.1)	Kersting and Van Wijngaarden 1992
Crawling water beetle *Petodytes* sp.	adult	S,U	Tech(94%)	N/A	96-h	LC_{50}	0.8 (N/A)	Federle and Collins 1976
Crayfish *Orconectes immunis*	N/A	F,M	Tech(99.9%)	44.4 mg/L as $CaCO_3$	96-h	LC_{50}	6.0 (4.0-9.0)	Phipps and Holcombe 1985
Cutthroat trout *Oncorhynchus clarki*	N/A	S,U	Tech(97%)	44 mg/L as $CaCO_3$	96-h	LC_{50}	18.4 (15.6-21.7)	Mayer and Ellersieck 1986
Cutthroat trout *Oncorhynchus clarki*	N/A	S,U	Tech(97%)	44 mg/L as $CaCO_3$	96-h	LC_{50}	5.4 (4.1-7.1)	Mayer and Ellersieck 1986
Cutthroat trout *Oncorhynchus clarki*	N/A	S,U	Tech(97%)	162 mg/L as $CaCO_3$	96-h	LC_{50}	26.0 (16.8-40.3)	Mayer and Ellersieck
Cutthroat trout *Oncorhynchus clarki*	N/A	S,U	Tech(97%)	162 mg/L as $CaCO_3$	96-h	LC_{50}	13.4 (9.9-18.0)	Mayer and Ellersieck 1986
Eastern oyster *Crassostrea virginica*	<2-h	S,U	Tech(98%)	$20^{o}/_{oo}$	48-h	EC_{50}	1991 (1505-2809)	Borthwick and Walsh 1981
Fathead minnow *Pimephales promelas*	juv.	F,M	Tech(99.9%)	45.3 mg/L as $CaCO_3$	96-h	LC_{50}	203 (191-217)	Holcombe et al. 1982
Fathead minnow *Pimephales promelas*	larvae	F,M	Tech(99%)	45.8 mg/L as $CaCO_3$	96-h	LC_{50}	140 (120-160)	Jarvinen and Tanner 1982
Fathead minnow *Pimephales promelas*	N/A	F,M	Tech(99.9%)	44.4 mg/L as $CaCO_3$	96-h	LC_{50}	542 (225-1310)	Phipps and Holcombe 1985
Goldfish *Carassius auratus*	N/A	F,M	Tech(99%)	44.4 mg/L as $CaCO_3$	96-h	LC_{50}	>806 (N/A)	Phipps and Holcombe 1985

Table A-1. Continued -4-

Species	Life Stage[a]	Method[b]	Formulation	Salinity/ Hardness[a]	Test Length	Effect	Values (95% C.L.)[c]	Reference
Grass shrimp *Palaemonetes pugio*	juv.	F,U	Tech(92%)	26‰	48-h	EC_{50}	1.5 (N/A)	Mayer 1987
Gulf killifish *Fundulus grandis*	juv.	F,M	Tech(92%)	28‰	96-h	LC_{50}	1.8 (1.5-2.1)	Mayer 1987
Gulf toadfish *Opsanus beta*	juv.	F,M	Tech(92%)	25‰	96-h	LC_{50}	68 (N/A)	Mayer 1987
Gulf toadfish *Opsanus beta*	2 mo.	S,M	Tech(92%)	29-30‰	96-h	LC_{50}	520 (450-600)	Hansen et al. 1986
Inland silverside *Menidia beryllina*	juv. 72-d	F,M	Tech(92%)	N/A	96-h	LC_{50}	4.2 (3.4-5.4)	Clark et al. 1985
Lake trout *Salvelinus namaycush*	N/A	F,U	Tech(97%)	162 mg/L as $CaCO_3$	96-h	LC_{50}	244 (205.0-290.0)	Mayer and Ellersieck 1986
Longnose killifish *Fundulus similis*	adult	F,M	Tech(92%)	26‰	96-h	LC_{50}	4.1 (2.8-6.9)	Mayer 1987
Mysid *Mysidopsis bahia*	juv. 1-d	F,M	Tech(92%)	27‰	96-h	LC_{50}	0.035 (0.029-0.043)	Mayer 1987
Mysid *Mysidopsis bahia*	adult	F,U	Tech(92%)	27‰	96-h	LC_{50}	0.040 (0.030-0.043)	Mayer 1987
Mysid *Mysidopsis bahia*	juv.	S,U	Tech(98%)	20‰	96-h	LC_{50}	0.056 (0.032-0.10)	Borthwick and Walsh 1981
Mysid *Neomysis mercedis*	neonate	S,M	Tech(99%)	499 mg/L as $CaCO_3$	96-h	LC_{50}	0.16 (0.14-0.30)	CDFG 1992a
Mysid *Neomysis mercedis*	neonate	S,M	Tech(99%)	509 mg/L as $CaCO_3$	96-h	LC_{50}	0.14 (0.09-0.1825)	CDFG 1992a
Mysid *Neomysis mercedis*	neonate	S,M	Tech(99%)	515 mg/L as $CaCO_3$	96-h	LC_{50}	0.15 (0.09-0.1825)	CDFG 1992a
Pink shrimp *Penaeus duorarum*	juv.	F,U	Tech(92%)	26‰	48-h	EC_{50}	2.4 (N/A)	Mayer 1987
Rainbow trout *Oncorhynchus mykiss*	juv.	F,M	Tech(99.9%)	45.3 mg/L as $CaCO_3$	96-h	LC_{50}	8.0 (6.8-9.4)	Holcombe et al. 1982

Species	Life Stage[a]	Method[b]	Formulation	Salinity/ Hardness[a]	Test Length	Effect	Values (95% C.L.)[c]	Reference
Rainbow trout *Oncorhynchus mykiss*	N/A	S,U	Tech(97%)	N/A	96-h	LC_{50}	7.1 (6.0-8.4)	Macek et al. 1969
Rainbow trout *Oncorhynchus mykiss*	N/A	S,U	Tech (97%)	44 mg/L as $CaCO_3$	96-h	LC_{50}	51 (43-60)	Mayer and Ellersieck 1986
Sheepshead minnow *Cyprinodon variegatus*	adult	F,M	Tech (98%)	10°/∞	96-h	LC_{50}	140 (110-160)	Mayer 1987
Sheepshead minnow *Cyprinodon variegatus*	fry	S,U	Tech(98%)	20°/∞	96-h	LC_{50}	270 (235-309)	Borthwick and Walsh 1981
Snail *Aplexa hypnorum*	adult	F,M	Tech(99.9%)	44.4 mg/L as $CaCO_3$	96-h	LC_{50}	>806 (N/A)	Phipps and Holcombe 1985
Spot *Leiostomus xanthurus*	juv.	F,U	Tech (92%)	26°/∞	48-h	LC_{50}	7.0 (N/A)	Mayer 1987
Stonefly *Pteronarcys californica*	naiad	S,U	Tech(97%)	N/A	96-h	LC_{50}	10 (7-13)	Sanders and Cope 1968
Stonefly *Pteronarcella badia*	naiad	S,U	Tech(97%)	N/A	96-h	LC_{50}	0.38 (0.30-0.49)	Sanders and Cope 1968
Stonefly *Claassenia sabulosa*	naiad	S,U	Tech(97%)	N/A	96-h	LC_{50}	0.57 (0.39-0.83)	Sanders and Cope 1968
Striped mullet *Mugil cephalus*	juv.	F,M	Tech (92%)	25°/∞	96-h	LC_{50}	5.4 (4.0-6.9)	Mayer 1987
Tidewater silverside *Menidia peninsulae*	0-d	F,M	Tech(92%)	20°/∞	96-h	LC_{50}	1.0 (N/A)	Borthwick et al. 1985
Tidewater silverside *Menidia peninsulae*	7-d	F,M	Tech(92%)	20°/∞	96-h	LC_{50}	0.5 (0.5-0.6)	Borthwick et al. 1985
Tidewater silverside *Menidia peninsulae*	14-d	F,M	Tech(92%)	20°/∞	96-h	LC_{50}	0.4 (0.3-0.6)	Borthwick et al. 1985

Table A-1. Continued -6-

Species	Life Stage[a]	Method[b]	Formulation	Salinity/ Hardness[a]	Test Length	Effect	Values (95% C.L.)[c]	Reference
Tidewater silverside *Menidia peninsulae*	28-d	F,M	Tech(92%)	20°/∞	96-h	LC_{50}	0.9 (0.7-1.1)	Borthwick et al. 1985
Tidewater silverside *Menidia peninsulae*	juv.	F,M	Tech(-)	N/A	96-h	LC_{50}	1.3 (1.0-1.7)	Clark et al. 1985
Tidewater silverside *Menidia peninsulae*	larvae 1-d	F,M	Tech(92%)	27°/∞	96-h	LC_{50}	0.96 (0.71-1.3)	Mayer 1987
Tidewater silverside *Menidia peninsulae*	larvae 7-d	F,M	Tech(92%)	20°/∞	96-h	LC_{50}	0.52 (0.46-0.59)	Mayer 1987
Tidewater silverside *Menidia peninsulae*	larvae 14-d	F,M	Tech(92%)	20°/∞	96-h	LC_{50}	0.42 (0.33-0.57)	Mayer 1987
Tidewater silverside *Menidia peninsulae*	larvae 28-d	F,M	Tech(92%)	30°/∞	96-h	LC_{50}	0.89 (0.69-1.1)	Mayer 1987

[a] N/A = Data not available

[b] F = Flow-through S = Static U = Unmeasured concentration M = Measured concentration

[c] 95% confidence limit

Table A-2. Values (μg/L) from unaccepted tests on the acute toxicity of chlorpyrifos to aquatic animals.

Species	Life Stage[a]	Method[a,b]	Formulation[a]	Salinity/ Hardness[a]	Test Length	Effect	Values (95% C.L.)[a,c]	Reference	Deficiencies[d]
Amphipod *Gammarus fasciatus*	N/A	S,N/A	Tech(97%)	N/A	96-h	LC_{50}	0.32 (0.12-0.90)	Sanders 1972	3
Atlantic silverside *Menidia menidia*	N/A	F,M	Tech(92%)	N/A	96-h	LC_{50}	1.7 (1.4-2.0)	Schimmel et al. 1983	3
Bluegill *Lepomis macrochirus*	N/A	S,N/A	N/A	2-5 mg/L as $CaCO_3$	96-h	LC_{50}	30 (N/A)	Carter and Graves 1973	3
Carp *Cyprinus carpio*	finger-ling	S,M	EC(40.8%)	116-123 mg/L as $CaCO_3$	48-h	LC_{50}	59 (38-92)	El-Refai et al. 1976	1,3,4,6
Carp *Cyprinus carpio*	finger-ling	S,M	EC(40.8%)	116-123 mg/L as $CaCO_3$	48-h	LC_{50}	280 (236-332)	El-Refai et al. 1976	1,3,4,6
Ceratopogonid *Culicoides variipennis*	4th instar	S,N/A	Tech(-)	N/A	24-h	LC_{50}	4.31 (3.8-4.9)	Holbrook 1982	1,3
Ceratopogonid *Culicoides variipennis*	larvae	S,U	Tech(-)	soft water	24-h	LC_{50}	2.96 (2.6-3.4)	Holbrook 1983	1,3
Channel catfish *Ictalurus punctatus*	N/A	S,N/A	N/A	2-5 mg/L as $CaCO_3$	24-h	LC_{50}	160 (N/A)	Carter and Graves 1973	3
Cladoceran *Ceriodaphnia dubia*	neonate	S,M	Tech(99%)	119 mg/L as $CaCO_3$	96-h	LC_{50}	0.12 (N/A)	CDFG 1992b	5
Cladoceran *Daphnia sp.*	adult & nymph	N/A	N/A	N/A	24-h	LC_{50}	16 (N/A)	Kenaga et al. 1965	3
Coral *Pocillopora damicornis*	planulae	S,F,N/A	Tech(-)	N/A	96-h	LC_{50}	N/A	Acevedo 1991	3
Crayfish *Procambarus acutus*	N/A	S,N/A	2-5 mg/L	N/A	96-h	LC_{50}	2.0 (N/A)	Carter and Graves 1973	3
Crayfish *Procambarus clarkii*	N/A	S,N/A	Tech(99.8%)	250 mg/L as $CaCO_3$	96-h	LC_{50}	21 (20-22)	Cebrian et al. 1992	3

Table A-2. Continued -2-

Species	Life Stage[a]	Method[a,b]	Formulation[a]	Salinity/ Hardness[a]	Test Length	Effect	Values (95% C.L.)[c]	Reference	Deficiencies[d]
European eel *Anguilla anguilla*	N/A	F,N/A	Tech(97%)	250 mg/L as $CaCO_3$	96-h	LC_{50}	540 (420-650)	Ferrando et al. 1991	2,3
Golden shiner *Notemigonus osysoleucus*	N/A	N/A	Tech(99±1%)	24 mg/L as $CaCO_3$	36-h	LC_{50}	35 (N/A)	Ferguson et al. 1966	1,7
Goldfish *Carrassius auratus*	juv.	N/A	N/A	N/A	24-h	LC_{50}	180 (N/A)	Kenaga et al. 1965	1,3
Green sunfish *Lepomis cyanellus*	N/A	N/A	Tech(99±1%)	24 mg/L as $CaCO_3$	36-h	LC_{50}	22.5 (N/A)	Ferguson et al. 1966	1,7
Green sunfish *Lepomis cyanellus*	adult	S,N/A	N/A	N/A	72-h	LC_{50}	40 (30-50)	Davey et al. 1976	1,3,9
Korean shrimp *Palaemon macrodactylus*	N/A	S,N/A	Tech(99%)	15-30‰	96-h	LC_{50}	0.25 (0.10-0.63)	Earnest 1970	3
Korean shrimp *Palaemon macrodactylus*	N/A	F,N/A	Tech(99%)	15-30‰	96-h	TL_{50}	0.01 (0.002-0.046)	Earnest 1970	3
Longnose killifish *Fundulus similis*	N/A	F,N/A	Tech(92%)	N/A	96-h	LC_{50}	4.1 (2.8-6.9)	Schimmel et al. 1983	3
Midge *Chironomus salinarius*	4th instar	N/A, N/A	Tech(-)	27-45‰	24-h	LC_{50}	0.44 (0.3-0.6)	Ali and Majori 1982	2,3,7
Mosquito *Wyeomyia smithii*	2nd instar	S,N/A	Tech (93-100%)	N/A	7-d	LC_{50}	N/A	Strickman 1985	3
Mosquito *Culicoides pipiens quinquefasciatus*	1st instar	N/A	Tech(-)	N/A	N/A	EC_{50}	N/A	U.S. Army Environ. Hygiene Agency 1970	3

Table A-2. Continued -3-

Species	Life Stage[a]	Method[a,b]	Formulation[a]	Salinity/ Hardness[a]	Test Length	Effect	Values (95% C.L.)[c]	Reference	Deficiencies[d]
Mosquitofish *Gambusia affinis*	N/A	N/A	Tech(99±1%)	24 mg/L as $CaCO_3$	36-h	LC_{50}	215 (N/A)	Ferguson et al. 1966	1,7
Mosquitofish *Gambusia affinis*	N/A	S,N/A	N/A	2-5 mg/L as $CaCO_3$	96-h	LC_{50}	280 (N/A)	Carter and Graves 1973	3
Mosquitofish *Gambusia affinis*	N/A	N/A	Tech(-)	N/A	92-h	LC_{85}	1000 (N/A)	Darwazeh and Mulla 1974	1,3
Mosquitofish *Gambusia affinis*	adult (gravid)	S,N/A	N/A	N/A	72-h	LC_{50}	260 (180-340)	Davey et al. 1976	1,3,9
Mummichog *Fundulus heteroclitus*	N/A	F,N/A	Tech(99.5%)	20-25‰	96-h	LC_{50}	4.65 (3.4-6.3)	Thirugnanum and Forgash 1977	3
Mysid *Mysidopsis bahia*	N/A	F,N/A	Tech(92%)	N/A	96-h	LC_{50}	0.035 (0.029-0.043)	Schimmel et al. 1983	3
Rotifer *Brachionus calyciflorus*	newly hatched	S,N/A	N/A	80-100 mg/L as $CaCO_3$	24-h	LC_{50}	11850 (10860-12840)	Ferrando and Andreu-Moliner 1991	3
Rotifer *Brachionus plicatilis*	newly hatched	S,N/A	N/A	15‰	24-h	LC_{50}	10670 (9310-12030)	Ferrando and Andreu-Moliner 1991	3
Sheepshead minnow *Cyprinodon variegatus*	N/A	F,N/A	Tech(92%)	N/A	96-h	LC_{50}	136 (113-153)	Schimmel et al. 1983	3
Snail *Helisoma trivolvis*	mature	N/A	N/A	N/A	24-h	LC_{50}	>2000 (N/A)	Kenaga et al. 1965	1,3
Striped bass *Morone saxatilis*	N/A	F,N/A	Tech(90%)	15-30‰	96-h	TL_{50}	0.58 (0.35-0.97)	Earnest 1970	3
Striped mullet *Mugil cephalus*	N/A	F,N/A	Tech(92%)	N/A	96-h	LC_{50}	5.4 (4.0-6.9)	Schimmel et al. 1983	3
Tadpole shrimp *Triops longicaudatus*	4-5 d	S,N/A	N/A	N/A	24-h	LC_{50}	4.0 (1.7-4.9)	Walton et al. 1990	3
Tilapia *Tilapia nilotica*	finger-ling	S,M	Tech(41.%)	116-123 mg/L as $CaCO_3$	48-h	LC_{50}	62 (42-91)	El-Rafai et al. 1976	1,3,4,6

Table A-2. Continued -4-

Species	Life Stage[a]	Method[a,b]	Formulation[a]	Salinity/ Hardness[a]	Test Length	Effect	Values (95% C.L.)[c]	Reference	Deficiencies[d]
Tilapia *Tilapia nilotica*	finger-ling	S,M	Tech(41%)	116-123 mg/L as $CaCO_3$	48-h	LC_{50}	114 (84-156)	El-Rafai et al. 1976	1,3,4,6

[a] N/A = not available

[b] S = Static F = Flow-through M = Measured concentration U = Unmeasured concentration

[c] Confidence limits

[d] 1 = Test duration too short or too long
6 = Pesticide formulation too low in active ingredient
2 = Unacceptable or unmeasured dissolved oxygen
7 = Test organisms exposed to pesticides prior to testing
3 = Essential information lacking 8 = Unacceptable dilution water used
4 = Test temperature varied by >2°C
9 = Inadequate number of concentrations tested
5 = Unacceptable control survival 10 = Unacceptable mortality range

APPENDIX B. Abstracts of chronic toxicity tests.

Accepted chronic toxicity tests - The following tests used accepted test methods.

Cripe et al. (1986) - In 1986, 28-d flow-through chronic toxicity tests were performed by the U.S. EPA on technical grade chlorpyrifos (92%) with sheepshead minnow *Cyprinodon variegatus*. Commonly recognized chronic test guidelines were not mentioned. Five concentrations were tested and a solvent control was used. Two replicates per concentration were tested. Chlorpyrifos concentrations were measured weekly and averaged 66 to 76% of nominal concentrations for the first series of tests and 64 to 74% of nominal concentrations for the second series of tests. Water quality parameters during the test averaged: temperature of 30 $\pm$1°C; pH range of 7.9 to 8.1; dissolved oxygen level of 5.0 mg/L; and salinity of 28°/₀₀ for the first series of tests and 25°/₀₀ for the second series of tests. Control survival was 97 to 100% for the first series of tests and 96 to 100% for the second series of tests. The NOEC value and LOEC value, based on growth, for *C. variegatus* were 1.7 µg/L and 3.0 µg/L, respectively. The MATC value was 2.26.

Goodman et al. (1985a) - In 1985, 28-d flow-through toxicity tests were performed by the U.S. EPA on technical grade chlorpyrifos (97.7%) with 32 to 36-h old embryos of inland silverside *Menidia beryllina* and tidewater silverside *Menidia peninsulae*. Commonly recognized testing guidelines were not mentioned. Five concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. One replicate per concentration was tested. Chlorpyrifos concentrations were measured weekly and averaged 63 to 90% of nominal concentrations in *M. beryllina* tests and 65 to 78% in *M. peninsulae* tests. Water quality parameters during the test averaged: temperature of 25 $\pm$2°C; pH was not measured but was estimated to range from 7.3

to 8.1; dissolved oxygen level of 7.6 mg/L to 7.8 mg/L for *M. beryllina* and 6.0 mg/L to 6.9 mg/L for *M. peninsulae*; and salinity of 4.0°/₀₀ to 6.0°/₀₀ for *M. beryllina* and 18°/₀₀ to 25°/₀₀ for *M. peninsulae*. Sea water and solvent control survival for *M. beryllina* were 80% and 83%, respectively. Sea water and solvent control survival for *M. peninsulae* were 88% and 63%, respectively. NOEC values, based on growth, for *M. beryllina* and *M. peninsulae* were 0.75 µg/L and 0.38 µg/L, respectively. The LOEC values based on growth, for *M. beryllina* and *M. peninsulae* were 1.8 µg/L and 0.78 µg/L, respectively. The MATC values for *M. beryllina* and *M. peninsulae* were 1.16 and 0.54, respectively. This test also included Atlantic silverside *Menidia menidia,* described in ***Unaccepted chronic toxicity tests***.

<u>Hansen et al. (1986)</u> - In 1986, a 49-d flow-through toxicity test was performed by the U.S. EPA on technical chlorpyrifos (92%) with gulf toadfish *Opsanus beta*. ASTM (1985) testing guidelines were followed. Six concentrations of chlorpyrifos were tested and a solvent control was used. Two to three replicates per concentration were tested. Chlorpyrifos concentrations were measured weekly and averaged 50 to 60% of nominal concentrations. Water quality parameters during the test averaged: temperature of 26 ±2°C; pH was not mentioned; dissolved oxygen level of 4.1 mg/L to 6.4 mg/L; and salinity of 25°/₀₀ to 34.5°/₀₀. Control survival was 97% The NOEC value and LOEC value, based on growth, were 1.4 µg/L and 3.7 µg/L, respectively. The MATC value for *O. beta* was 2.28.

<u>Jarvinen and Tanner (1982)</u> - In 1982, 32-d flow-through toxicity tests were performed by the U.S. EPA on technical grade chlorpyrifos (98.7%) with <48-h embryos of fathead minnow *Pimephales promelas*. APHA (1975) testing guidelines were followed. Five concentrations of chlorpyrifos were tested and a dilution water control was used. Two replicates per

concentration were tested. Chlorpyrifos concentrations were measured weekly and averaged 102 to 144% of nominal concentrations. Water quality parameters during the test averaged: temperature of 25.0 ±0.6°C; pH of 7.4 to 7.8; dissolved oxygen level of 6.5 mg/L to 8.4 mg/L; hardness of 45.8 mg/L; and alkalinity of 43.1 mg/L. Control survival was 100%. The NOEC value and LOEC value, based on survival, for *P. promelas* were 1.6 µg/L and 3.2 µg/L, respectively. The MATC value for *P. promelas* was 2.26.

McKenney et al. (1981) - In 1981, 28-d flow-through toxicity tests were performed by the U.S. EPA on technical grade chlorpyrifos (97.7%) with juvenile mysid *Mysidopsis bahia*. Commonly recognized testing guidelines were not mentioned. Four concentrations of chlorpyrifos were tested and a solvent control was used. Eight replicates per concentration were tested. Chlorpyrifos concentrations were measured weekly and averaged 71 to 120% of nominal concentrations. Water quality parameters during the test averaged: temperature of 25 ±2°C; pH was not measured; dissolved oxygen level of 6.6 ±0.2 mg/L; and salinity of 19‰ to 28‰. Control survival was 74%. The NOEC value, LOEC value, and MATC value, based on growth, for *M. bahia* were 0.002 µg/L, 0.004, and 0.003 µg/L, respectively.

Norberg and Mount (1985) - In 1985, 7-d static toxicity tests were performed by U.S. EPA on technical Dursban® (percent active ingredient not specified) with larval fathead minnow *Pimephales promelas*. EPA (1982) and ASTM (1983) testing guidelines were followed. Five concentrations were tested and a dilution water control was used. Three replicates per concentration were tested. Chlorpyrifos concentrations were measured and averaged 60 to 74% of nominal concentrations. Water quality parameters during the test averaged: temperature of 25 ±2°C; pH of 7.8 to 8.0; initial dissolved oxygen level of 8.0 mg/L to 6.0 mg/L at

24-h; and hardness of 45 mg/L to 48 mg/L as $CaCO_3$. Control survival was ≥80%. The 7-d NOEC value and LOEC value for *P. promelas* based on growth were 3.7 µg/L and 7.4 µg/L, respectively. The MATC value was 5.23.

Unaccepted chronic toxicity tests - The following tests did not use accepted test methods.

Goodman et al. (1985b) - In 1985, 26-d and 35-d flow-through toxicity tests were performed by the U.S. EPA on technical grade chlorpyrifos (92%) with 2.5-d old fry California grunion *Leuresthes tenuis*. Commonly recognized testing guidelines were not mentioned. Five concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. Three replicates per concentration were tested. Chlorpyrifos concentrations were nominal. Water quality parameters during the test averaged: temperature of 23°C to 26°C; pH of 7.6 to 7.9; dissolved oxygen level of 5.7 mg/L to 5.8 mg/L; and salinity of 28.6‰ in the 35-d tests and 29.3‰ in the 29-d tests. Control survival was 85% in the 26-d toxicity test and 78 to 82% in the 35-d toxicity test. The 26-d NOEC value and LOEC values for *L. tenuis* were 0.50 µg/L and 1.0 µg/L, respectively. The 35-d NOEC value and LOEC value for *L. tenuis* were 0.25 µg/L and 0.50 µg/L, respectively. These values were not used because chlorpyrifos concentrations were nominal and 2.5-d old fry were used instead of <48-h old embryos.

Goodman et al. (1985a) - In 1985, 28-d flow-through toxicity tests were performed by the U.S. EPA on technical grade chlorpyrifos (92%) with 32 to 36-h old embryos of Atlantic silverside *Menidia menidia*. Commonly recognized testing guidelines were not mentioned. Five concentrations of chlorpyrifos were tested and a solvent control was used. One replicate per concentration was tested. Chlorpyrifos concentrations were measured weekly and averaged 48 to 132% of nominal concentrations. Water quality parameters during the test averaged: temperature of 25 $\pm 2^{\circ}C$; pH was not mentioned; dissolved oxygen level of 5.2 mg/L to 5.5 mg/L; and a salinity of $18^{\circ}/_{\circ\circ}$ to $27^{\circ}/_{\circ\circ}$. Control survival was 41%. The NOEC value and LOEC value for *M. menidia* were 0.28 µg/L and 0.48 µg/L, respectively. These data were not used because control survival in chronic toxicity tests must be greater than 60% (ASTM 1988c). The *M. menidia* tests had a control survival of 41%. This test also included tests using inland silverside *Menidia beryllina* and tidewater silverside *Menidia peninsulae*. These tests were acceptable and are described in ***Acceptable chronic toxicity test abstracts***.

Kersting and Van Wijngaarden (1992) - In 1992, 21-d static toxicity tests were performed by the Research Institute for Nature Management, Netherlands on technical grade chlorpyrifos (99%) with <24-h old cladoceran *Daphnia magna*. Commonly recognized testing guidelines were not mentioned. Six concentrations of chlorpyrifos were tested and solvent and dilution water controls were used. Two replicates per concentration were tested. Chlorpyrifos concentrations were measured at the beginning of each test and averaged 60% of nominal concentrations. Water quality parameters during the test averaged: temperature of 19.5 $\pm 0.5^{\circ}C$; pH of 6.8 to 7.0; and dissolved oxygen level of 7.7 mg/L to 8.8 mg/L. Control survival was 100%. The NOEC value, based on reproduction, for *D. magna* was 0.1 µg/L. These values were not used because a LOEC value was not determined and concentrations were not measured during the test.

Table B-1. Values (μg/L) from accepted tests on the chronic toxicity of chlorpyrifos to aquatic animals.

Species	Life Stage[a]	Method[b]	Formulation	Salinity/ Hardness	Test Length	Effect	Values	MATC $(NOECXLOEC)^{1/2}$	Reference
Fathead minnow *Pimephales promelas*	embryo	F,M	Tech(99%)	45.8 mg/L as $CaCO_3$	32-d	NOEC LOEC	1.6 3.2	2.26	Jarvinen and Tanner 1982
Fathead minnow *Pimephales promelas*	larvae	S,M	Tech(--)	45-48 mg/L as $CaCO_3$	7-d	NOEC LOEC	3.7 7.4	5.23	Norberg and Mount 1985
Gulf toadfish *Opsanus beta*	embryo-juv.	F,M	Tech(92%)	25-34°/oo	49-d	NOEC LOEC	1.4 3.7	2.28	Hansen et al. 1986
Inland silverside *Menidia beryllina*	embryo	F,M	Tech(98%)	4-6°/oo	28-d	NOEC LOEC	0.75 1.80	1.16	Goodman et al. 1985a
Mysid *Mysidopsis bahia*	<48-h juv.	F,M	Tech(98%)	19-28°/oo	28-d	NOEC LOEC	0.002 0.004	0.003	McKenney et al. 1981
Sheepshead minnow *Cyprinodon variegatus*	embryo	F,M	Tech(92%)	25-28°/oo	28-d	NOEC LOEC	1.7 3.0	2.26	Cripe et al. 1986
Tidewater silverside *Menidia peninsulae*	embryo	F,M	Tech(98%)	18-25°/oo	28-d	NOEC LOEC	0.38 0.78	0.544	Goodman et al. 1985a

[a] N/A = Data not available

[b] S = Static   F = Flow-through   M = Measured concentration   U = Unmeasured concentration

Table B-2. Values (µg/L) from unaccepted tests on the chronic toxicity of chlorpyrifos to aquatic animals.

Species	Life Stage	Method[a]	Formulation	Salinity/ Hardness[b]	Test Length	Effect	Values[b]	Reference	Deficiencies[c]
Atlantic silverside *Menidia menidia*	embryo	F,M	Tech(92%)	18-27°/₀₀	28-d	NOEC LOEC	0.28 0.48	Goodman et al. 1985a	4
California grunion *Leuresthes tenuis*	fry	F,U	Tech(92%)	29.3°/₀₀	26-d	NOEC LOEC	0.5 1.0	Goodman et al. 1985b	5,6
California grunion *Leuresthes tenuis*	2.5-d embryo	F,U	Tech(92%)	28.6°/₀₀	35-d	NOEC LOEC	0.25 0.50	Goodman et al. 1985b	5,6
Cladoceran *Daphnia magna*	<24-h	S,U	Tech(99%)	N/A	21-d	NOEC LOEC	0.1 N/A	Kersting and Van Wijngaarden 1992	1,5

[a] F = Flow-through S= Static M = Measured concentration U = Unmeasured concentration

[b] N/A = Data not available

[c] 1 = Essential information lacking
2 = Unacceptable or unmeasured dissolved oxygen levels
3 = Test organisms exposed to pesticides prior to testing
4 = Unacceptable control survival
5 = Concentrations not measured
6 = Unacceptable life stage tested

APPENDIX C. Abstracts of plant toxicity tests.

Borthwick and Walsh (1981) - In 1981, 96-h static toxicity tests were performed by the U.S. EPA on technical grade Dursban^R (97%) with the diatoms *Skeletonema costatum* (5 tests), *Isochrysis galbana* (1 test), and *Thalassiosira pseudonana* (1 test). ASTM (1978a,b) testing guidelines were followed. Five concentrations of Dursban^R were tested and solvent and dilution water controls were used. Two replicates per concentration were tested. Measurement of chlorpyrifos concentrations was not mentioned for any test. Water quality parameters during the test averaged: temperature of 20.0 $\pm$0.5°C; pH of 8.1; dissolved oxygen level was not measured; and salinity of 30°/$_{oo}$. The 96-h EC_{50} values for *S. costatum* were 255 µg/L, 289 µg/L, 326 µg/L, 328 µg/L, and 297 µg/L for tests 1-5, respectively. The 96-h EC_{50} values, based on growth inhibition, for *I. galbana* and *T. pseudonana* were 138 µg/L and 148 µg/L, respectively. This test also included tests with mysid *Mysidopsis bahia*, eastern oyster *Crassostrea virginica*, and sheepshead minnow *Cyprinodon variegatus* (Appendix A).

Brown et al. (1976) - In 1973-74, static toxicity tests were performed by the Institute for Environmental Studies and School of Hygiene, University of Toronto on technical grade Dursban^R (percent active ingredient not specified) with phytoplankton. The commonly occurring species of phytoplankton in the artificial pond system were: *Ankistrodesmus* sp., *Ceratium* sp., *Closteriopsis* sp., *Dinobyron* sp., *Glenodinium* sp., *Tetraedron* sp., and diatoms. Commonly recognized testing guidelines were not mentioned. Four concentrations of Dursban^R were tested and a solvent control was used. The number of replicates tested was not mentioned. Dursban^R was applied once and there was a decline in the concentration of the insecticide over time. Water quality parameters during the test were not mentioned. Critical values were not reported.

Butcher et al. (1977) - In 1977, static toxicity tests longer than 80-d were performed by the Department of Biology and Centre for Research on Environmental Quality, York University, Canada on Dursban® (percent active ingredient not specified) with green and blue-green algae (major species: *Mougeotia* sp., *Chlorella* sp.). Commonly recognized testing guidelines were not mentioned. Three concentrations of Dursban® were tested and water controls were used. Only one treatment per concentration was tested. Chlorpyrifos concentrations tested were nominal. Water quality parameters during the test averaged: temperature of 20°C to 25°C; pH was not mentioned; and dissolved oxygen level of 7.3 mg/L to 10.7 mg/L; hardness/salinity was not mentioned. Algal blooms in the control ponds were significantly smaller and less persistent than the blooms observed in treated ponds. Zooplankton in treated ponds were almost completely eliminated 48-h after chlorpyrifos exposure.

Lal et al. (1987) - In 1987, 35-d growth studies and 5-d nitrogen fixation studies were performed by the Department of Zoology, Sri Venkateswara College, India on technical grade chlorpyrifos (>96%) with blue green algae *Anabaena* and *Aulosira fertilissima*. Commonly recognized testing guidelines were not mentioned. Three concentrations of chlorpyrifos were tested and a solvent control was used. Two replicates per concentration were tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 28°C to 30°C; pH of 7.5; dissolved oxygen level was not mentioned; and salinity not mentioned. At 5 mg/L and 10 mg/L chlorpyrifos was found to generate morphological effects on cellular orientation and number within algal filaments in both species of algae. A slight stimulation in growth was observed in *Aulosira fertillissima* at 1 mg/L chlorpyrifos (the lowest concentration tested). Growth inhibition was observed for the other concentrations of chlorpyrifos tested with *Aulosira* *fertillissima*. Growth inhibition was observed in *Anabaena* at all concentrations of chlorpyrifos tested.

Maly and Ruber (1983) - In 1983, 96-h static toxicity tests were performed by the Department of Biology, Northeastern University, Boston on technical grade chlorpyrifos (percent active ingredient not specified) with diatoms *Nitzschia closterium*, *Amphora coffeaformis v. borealis*, and *Amphiprora* sp., the green algae *Chlorococcum* sp., and dinoflagellate *Gonyaulax* sp. Commonly recognized testing guidelines were not mentioned. Four concentrations of chlorpyrifos were tested and a solvent control was used. The number of replicates tested was not mentioned. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 23 $\pm$2°C to 32 $\pm$3°C; pH was not mentioned; and salinity of 27°/$_{oo}$. Control survival was not mentioned. Critical values were not reported.

Steelman et al. (1966) - In 1966, 24-h and 48-h toxicity tests were performed by Louisiana State University on Dursban^R (percent active ingredient not specified) with coliform, amylolytic, enterococci, lipolytic, and proteolytic bacteria. Society of American Bacteriologists (1957) testing guidelines were followed. Five concentrations of Dursban^R were tested and a water control was used. Three replicates per concentration were tested. Chlorpyrifos concentrations tested were nominal. Water quality parameters during the test averaged: temperature was not mentioned; pH of 7.0; the medium was aerated; and hardness was not mentioned. There was about 24 million bacteria cultures in the control at 24-h and approximately 29 million at 48-h. Dursban^R was found to have a small effect on bacterial populations. The LC_{50} value for the bacteria was 0.028% Dursban^R. Important information, such as the percent active ingredient, was lacking.

Walsh (1983) - In 1983, 48-h static toxicity tests were performed by the U.S. EPA on chlorpyrifos (the percent active ingredient not specified) with diatom *Skeletonema costatum*. APHA (1975) testing guidelines were followed. Five concentrations of chlorpyrifos were tested and a solvent control was used. Three replicates per concentration were tested. Measurement of chlorpyrifos concentrations was not mentioned. Water quality parameters during the test averaged: temperature of 20 $\pm$0.5°C; pH of 8.1; dissolved oxygen level not mentioned; and salinity of 30°/₀₀. Control survival was 96%. The 48-h EC_{50} value, based on growth, for *S. costatum* was 1200 µg/L. Important information, such as percent active ingredient, was lacking.

APPENDIX D. Procedures used by the California Department of Fish and Game to prepare hazard assessments.

The California Department of Fish and Game (CDFG) Pesticide Investigations Unit assesses the hazard of pesticides to aquatic organisms. The hazard assessment procedure includes evaluation of toxicity studies, establishment of Water Quality Criteria (WQC), and assessment of potential hazards.

Acute and chronic toxicity data are obtained from studies published in scientific literature and laboratory reports required by EPA for pesticide registration. The CDFG evaluates the quality of these data by evaluating the tests for compliance with standards adapted from the Environmental Protection Agency (EPA) and the American Society for Testing and Materials (ASTM). The tests are evaluated for compliance with standards for test type, method, design and species, and for water quality and toxicant monitoring and maintenance. Although a study need not comply with every standard, tests are rejected if they do not observe certain fundamental procedures or if several important standards are not met. Studies are also rejected if they do not contain sufficient information to be properly evaluated and the necessary information cannot be obtained from the original researcher.

Acute toxicity data from acceptable tests on freshwater and saltwater organisms are used to determine a Final Acute Value (FAV). The EPA (1985) guidelines recommend eight families of freshwater organisms for which data should be available for deriving a freshwater FAV, and eight families of saltwater organisms for deriving a saltwater FAV. The EPA (1985) procedure does not discuss derivation of an estuarine FAV. The FAV is calculated as follows:

1. The Species Mean Acute Value (SMAV) is the geometric mean of EC_{50} values and LC_{50} values from all accepted toxicity tests performed on that species.

2. The Genus Mean Acute Value (GMAV) is the geometric mean of all SMAVs for each genus.

3. The GMAVs are ranked (R) from "1" for the lowest to "N" for the highest. Identical GMAVs are arbitrarily assigned successive ranks.

4. The cumulative probability (P) is calculated for each GMAV as R/(N+1).

5. The four GMAVs with cumulative probabilities closest to 0.05 are selected. If fewer than 59 GMAVs are available, these will always be the four lowest GMAVs.

6. The FAV is calculated using the selected GMAVs and Ps, as follows:

$$S^2 = \frac{\Sigma((\ln GMAV)^2) - ((\Sigma(\ln GMAV))^2/4)}{\Sigma(P) - ((\Sigma(\sqrt{P}))^2/4)}$$

$$L = (\Sigma(\ln GMAV) - S(\Sigma(\sqrt{P})))/4$$

$$A = S(\sqrt{0.05}) + L$$

$$FAV = e^A$$

Chronic toxicity data from acceptable tests on freshwater and saltwater organisms are used to determine a Final Chronic Value (FCV). If data are available for the eight freshwater and saltwater families, the FCV is calculated using the same procedure as described for the FAV. If sufficient data are not available, the following procedure is used:

1. Chronic values are obtained by calculating the geometric mean of the NOEC value and the LOEC value from acceptable chronic toxicity tests.

2. Acute-Chronic Ratios (ACR) are calculated for each chronic value for which at least one corresponding acute value is available. Whenever possible, the acute test(s) should be part of the same study as the chronic test.

3. The Final ACR value (FACR) is calculated as the geometric mean of all the species mean ACR values available for both freshwater and saltwater species.

4. FCV = FAV/FACR.

Plant toxicity data from algae or aquatic vascular plants are used to determine a Final Plant Value (FPV). The FPV is the lowest result from a test with a biologically important endpoint.

The WQC is equivalent to the lowest of three values: FAV, FCV, or FPV. Separate WQCs can be generated for freshwater and saltwater species if the specific water system is solely saltwater or freshwater. The WQC can be lowered to protect important species.

The CDFG compares the WQC with concentrations detected in specific waters. If concentrations are greater than the WQC, the

CDFG determines that aquatic organisms may be threatened, and potential solutions are explored.

Hazard assessment is an iterative process by which new data are evaluated to refine the WQC. Hazard assessments frequently recommend additional toxicity tests with sensitive native species and commonly-used testing organisms listed by ASTM.